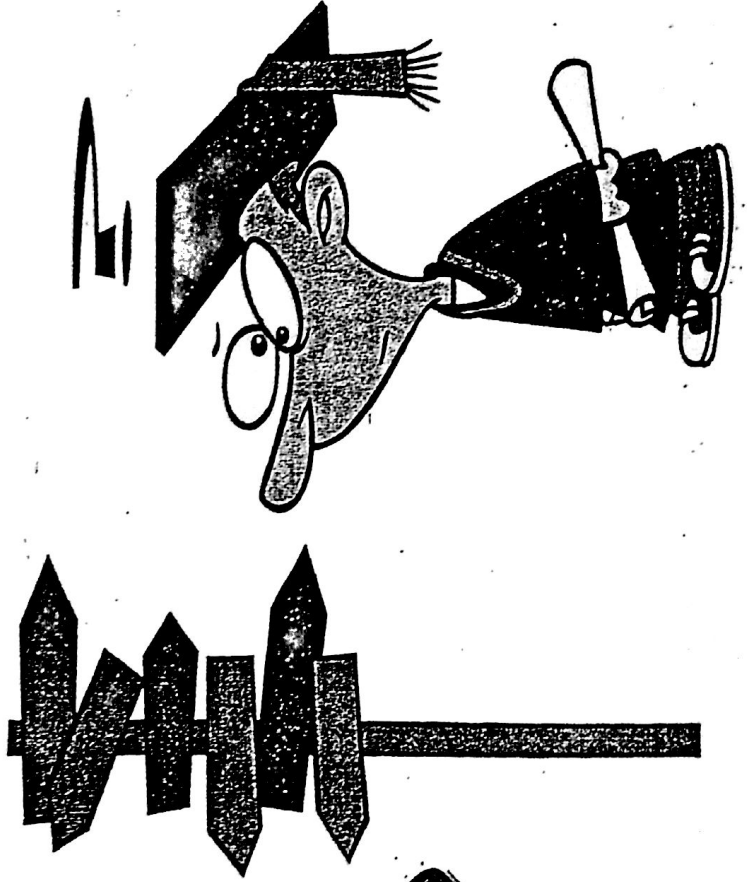


Systematic Bacteriology (Exams Guide)

سراجو بكتريا
حلول امتحانات
+ تجربات بكتريا

حلول امتحانات الأعوام السابقة (٢٠٠٨ - ٢٠١٤)

COLLECTIONS

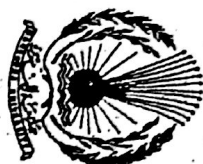


Please, follow up the questions in back of your questions paper

MANSOURA UNIVERSITY

FACULTY OF VETERINARY MEDICINE

DEPT. BACTERIOLOGY, MYCOLOGY AND IMMUNOLOGY



SYSTEMATIC BACTERIOLOGY EXAM.
3rd year Students, 2nd Semester (2013/2014)

Exam. Date 6/11/2014

Answer the following questions Full mark: 25 Time: 2 hours

1) Comment on (Answer only 3 points): (6 marks)

- 1- Serodiagnosis of brucellosis in cattle.
- 2- General characters of mycoplasmas.
- 3- Mechanism of invasion of cells and intracellular spread by *L. monocytogenes*.
- 4- Antigenic structure of *P. multocida*.

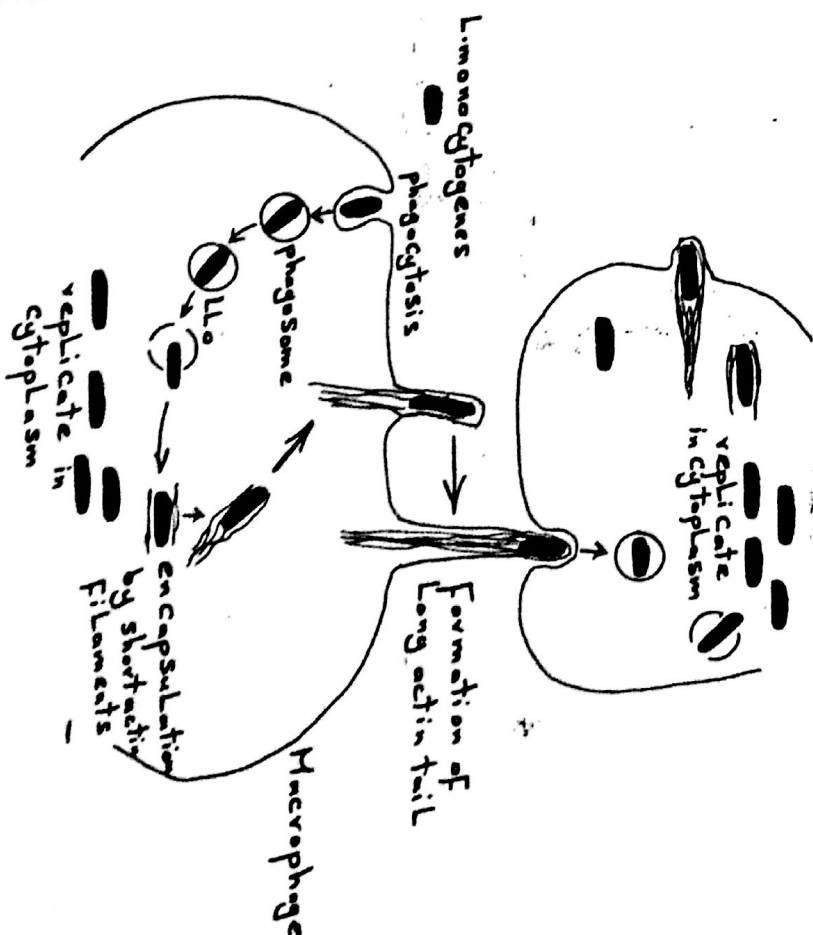
① Serodiagnosis of brucellosis in cattle
C.11 ٥٤٥١ ٥٤٥١

② General characters of mycoplasmas
C.11 ٥٤٥١ ٥٤٥١

③ Mechanism of invasion of cells and intracellular spread by *L. monocytogenes*

L. monocytogenes induce phagocytosis through a membrane associated protein called internalin → once ingested,

The bacterium produce Listeriolysin (LLO) to escape from the phagosome → then multiply rapidly in the cytoplasm → move through the cytoplasm to invade adjacent cells by polymerizing actin to form long tails.



④ antigenic structure of *P. multocida*

C.11 ٥٤٥١ ٥٤٥١

1

2) Discuss (Answer only 3 points):

(6 marks)

1. Runyon's classification of Mycobacteria.
2. Different types of classification of Clostridia.
3. Vaccination against *Pasteurella multocida* infection in buffaloes.
4. Immunization against *B. anthracis*.

① Runyon's classification of Mycobacteria

Mycobacteria is classified into 4 groups acc. to rate of growth and pigment production:

a- slow growers:

1- group I (photochromogenic):

- produce yellow pigmented colonies only after exposure to light.
- slow growing (require 7 days or ↑ for visible growth to be seen)
e.g. *M. Kansasii* and *M. marinum*

2- group II (scotochromogenic):

- produce yellow or orange pigment either in the presence or absence of light.
- slow growing
e.g. *M. goodii*, *M. xenopi* and *M. scrofulaceum*.

3- group III (Non-photochromogenic):

- produce no or slight pigment with exposure to light
- slow growing
e.g. *M. ulcerans*

b- Rapid growers:

group IV (variable pigmentation):

- give variable pigmentation
- grow rapidly (visible growth is seen in less than 7 days).
e.g. *M. fortuitum*, *M. phlei* and *M. smegmatis*

② different types of classification of clostridia

C.IV اكل الجلل

③ Vaccination against *P. multocida* infection in buffaloes.

C.II اكل الجلل

④ Immunization against *B. anthracis*

C.IV اكل الجلل

2

What are the virulence factors of.....?

(3 marks)

1. *Pseudomonas aeruginosa*.
2. Members of *F. Enterobacteriaceae*.

① Virulence Factors of *P. aeruginosa*

Cellular Components	Extracellular products
<p>1- pili</p> <p>2- Capsule (alginate)</p> <p>3- Lps (endotoxin)</p>	<p>1- proteases</p> <p>2- cytotoxin</p> <p>3- Hemolysins</p> <p>4- Enterotoxin</p> <p>5- Exotoxins</p> <p>6- Neuraminidase</p> <p>7- pigments</p>

A - Cellular Components:

pili	Capsule (alginate)	Lps (endotoxin)
<p>Facilitate adherence to epithelial cells with subsequent Colonization</p>	<p>poly saccharide in nature.</p> <p>act as antiphagocytic</p> <p>It is a mucoid alginate layer → Facilitate adherence to host cells.</p>	

B - Extracellular products:

1- Proteases: 2 types

Elastase

alkaline protease

- Cleave Collagen, IgG, IgA and Complement
- Lyse Fibrinectin to expose receptors for bacterial attachment on the mucosa of lung
- disrupt the resp. epithelium and interfere with ciliary function.
- Interfere with Fibrin formation and will lyse fibrin.
- Elastase and alkaline protease together:
 - 1- destroy the ground subs. of the cornea and other supporting structures composed of fibrin and elastin.
 - 2- Inactivate γ -IFN and TNF.

2 - Cytotoxin:

- a pore-forming protein
- was originally named Leukocidin → because of its effect on neutrophils, but it appears to be cytotoxic for most eukaryotic cells.

3 - Hemolysins

- They act synergistically to:
 - breakdown lipids and lecithin → necrosis
 - breakdown pulmonary surfactant with resulting atelectasis.

③

4- enterotoxin:
responsible for diarrhea during intestinal infection.

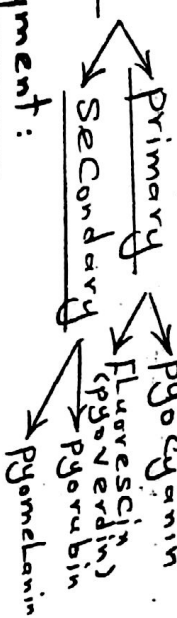
5- Exotoxins

<u>Exotoxin - A</u>	<u>Exotoxin - S</u>
toxic for monocytes and bone marrow	interfere with phagocytic killing

6- Neuraminidase:

remove sialic acid residues from the pili receptors → enhance adherence of the bacteria to epithelial cells.

7- pigments:



Pyocyanin pigment:

antimicrobial activities against a wide range of bacteria and some fungi →

So, it suppress the growth or kill other bacteria in mixed cultures → where it catalyze the reduced NAD → resulting in the conversion of O_2 to superoxide and H_2O_2 → inhibit the growth of other bacteria
→ cytotoxic to host cells.

mediate tissue damage → through the production of toxic oxygen radicals (H_2O_2 , superoxide and OH radical).

2 Members of F. enterobacteriaceae

C. 14 انتزاعا

4) What are the laboratory differences between.....? (3 marks)

1- α and β haemolysis of *S. aureus*.

2- *B. mallei* and *B. pseudomallei*.

3- Morphology of *P. multocida* and morphology of *Campylobacter* species.

① α and β - haemolysins of *S. aureus*

② *B. mallei* and *B. pseudomallei*

C. 14 انتزاعا

③ Morphology of *P. multocida* and morphology of *Campylobacter* species

Morphology	<i>P. multocida</i>	<i>Campylobacter</i> spp.
Stain used	Gram's stain	Gram's stain
staining reaction	Gram - ve	Gram - ve
Shape	Cocci/bacilli	Comma or S-shaped appear as flying saucer.
Capsule	Capsulated when recently isolated from disease condition	may be capsulated
Spore	Non	Non
Motility	Non-motile	motile with mono- or amphitrichous flagella. → motility is seen by phase contrast or dark ground illumination microscope → show characteristic darting movement (cystis' flying saucer organism)

④

Morphology of P. multocida

eli

- using Leishman's, Giemsa or methylene blue → bipolar staining (bipolarity)
- after repeated subculture on agar → the organism tend to become pleomorphic forming chains, filaments and rods of various sizes.

Please, follow up the questions in back of your questions paper

5) Why.....?

(2 marks)

- 1- Clostridia are obligatory anaerobic bacteria.
- 2- Mollicutes including mycoplasmas belong to phylum (division) Firmicutes.
- 3- The predilection site of brucellae is the reproductive tract of both male and female animals.
- 4- Brucellae are stained with modified Ziehl-Neelsen' stain.

① because they lack resp. enzymes (catalase, oxidase and peroxidase) → So, H_2O_2 will be formed → toxic to bacterial cell.

② because they have low GC-content (18 - 40 mol %) small genome.

5

③ because erythritol (which is essential for the growth of most brucellae) is present in the placenta and genital tract of cattle, sheep, goats and pigs.

④ because they resist decolorization by weak acids (such as 0.5% acetic acid) → So, they are stained red and other organisms stained blue by modified Ziehl-Neelsen's stain.

6) Put the mark (✓) or (X) in front of each of the following sentences and explain your answer (please, no marks without explain)

(5 marks)

1- Spores of *Staphylococcus aureus* are found subterminal. X

because *S. aureus* is non-spore forming bacteria (cocci).

2- Tetanospasmin is produced by both toxigenic and non-toxicogenic strains of *C. tetani*. X

because tetanospasmin is produced only by toxigenic strains of *C. tetani*.

3- Symptomatic anthrax is a disease infecting cattle and caused by

C. perfringens type A through ingestion of its spores. X

because symptomatic anthrax is a disease infecting sheep and caused by *C. chauvoei* through wound contamination by spore

4. Fowls are susceptible to infection by *C. chauvoei*. (X)
because Fowls have natural (innate) resistance against infection by *C. chauvoei*.

5. *Listeria* are halophilic organisms. (✓)
because *Listeria* can tolerate 10% NaCl.

6. Chinese letters and palisade arrangement is a character of salmonellae. (X)

Chinese letters and palisade arrangement is a character of *Corynebacteria* → because during division, the daughter cells remain attached in one side producing L and V arrangements that are referred to Chinese letters or arranged in rows parallel to each other giving palisade arrangement.

7. Ulcerative lymphangitis in horse is caused by *Streptococcus equi*. (X)
ulcerative lymphangitis in horse is caused by *Corynebacterium pseudotuberculosis* (*C. ovis*).

8. Cold enrichment is a character of *L. monocytogenes*. (✓)
Cold enrichment is necessary for *L. monocytogenes* → which is able to grow at refrigerator temperature
→ samples (brain or food samples) are homogenized → 10% suspension is made in broth → placed in refrigerator at 4°C → subcultured on solid media once weekly for up to 12 weeks.

9. Summer mastitis is caused by *S. agalactiae*. (X)
Summer mastitis is caused by → *peptostreptococcus indolicus* → *actinomyces pyogenes*.

10. Spoon like shape of spores is a character of *C. tetani*. (X)
drum-stick appearance of spore is a character of *C. tetani*.

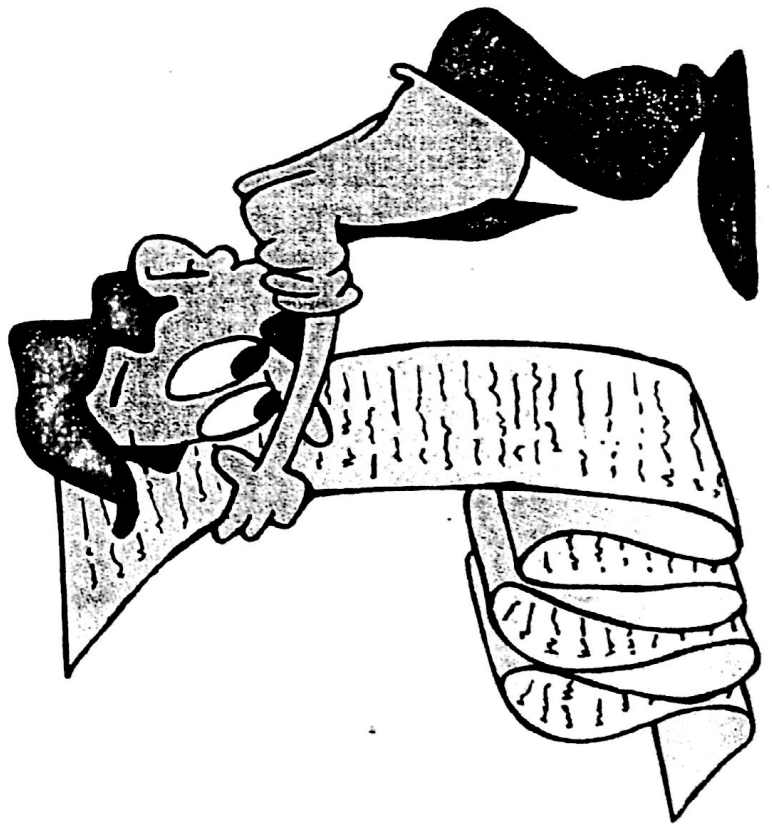
GOOD LUCK
Prof. Gamal Abd El-Gaber Mohamed Younis
Professor and Chairman of the Department

Please, follow up the questions in back of your questions paper

Systematic Bacteriology

(Exams Guide)

امتحان ۲۰۱۳



Bacteriology 2013

① correct the following sentences, explain your answer?

1- Mycoplasmas have usually fixed shape.
Mycoplasmas do not have fixed shape (pleomorphic) because they are devoid of cell wall peptidoglycan.

2- Mycoplasma colonies are seen by naked eyes.

Mycoplasma colonies can't be seen by naked eye → as Mycoplasma produce fried egg microcolonies which are the smallest colonies (ca. 1-0.6 mm in diameter) which are visible only under dissecting or stereo-microscope or low power of ordinary microscope or hand lens.

3- Digitonin test differentiates mycoplasma from ureaplasma.

Digitonin test differentiates mycoplasma from achleoplasma or modified urease test differentiates mycoplasma from ureaplasma.

4- Mycobacteria can be stained by Gram's stain.

Mycobacteria do not take up the dyes of Gram's stain because their cell walls are rich in lipids definitely mycolic acid, but they are stained by Ziehl Neelsen's (acid fast bacilli).

5- Mycoplasmas can be stained by Ziehl Neelsen's stain.

Mycoplasmas can't be stained by Ziehl Neelsen's stain because they lack cell wall.

6- BCG can be used for treatment of infected animals.

BCG vaccine (Bacille Calmette Guerin) is a vaccine used for immunization against Mycobacterium → It is a live bovine strain that is attenuated by growth in potato-glycerin bile salts medium through several hundred subcultures.

7- Tuberculin test can be used for diagnosis of Mycoplasma infection in poultry.

Tuberculin test is an allergic test used for diagnosis of Mycobacterium in animals and man (depend on delayed type of hypersensitivity)

8- Contagious bovine pleuropneumonia is the choice biochemical test used for diagnosis of Mycobacteria in poultry.

Contagious bovine pleuropneumonia (CBPP) is a disease caused by Mycoplasma mycoides subspecies mycoides in cattle.

g- pasteurellae can be easily grown on nutrient agar.

Pasteurellae fail to grow on ordinary media but, the growth is improved by the addition of blood, serum or 0.3% yeast extract.

10- Fowl cholera is the drug of choice for mycobacteria in cattle.

Fowl cholera is a disease in poultry caused by Pasteurella multocida (II: A: 5), II: A: 8 and II: A: 9).

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② What are the differences between:

1- α and B haemolysins of S. aureus.

	α haemolysin	B-haemolysin
Susceptible RBCs	Rabbit RBCs are the most susceptible	Lyse sheep RBCs but not rabbit RBCs.
Lethal to white mice	+	-
dermonecrosis	+	-
other characters	<ul style="list-style-type: none"> It is an artherius toxin \rightarrow i.e. it loses its activity at 70°C and retains its activity at 100°C due to dissociation Serve as a convenient index of virulence where the more virulent strain, the more α-hemolysin producer. Systemic release of toxin cause of septic shock. 	
	<ul style="list-style-type: none"> Produced most often by strains isolated from animal sources. 	

2- P. multocida and P. haemolytica

points of differences	P. multocida	H. haemolytica
① Haemolysis	—	B-haemolytic
② growth on MacConKey's agar	—	+
③ Lactose Fermentation	—	+(acid only)
④ Litmus milk	—	+(acid and clot)
⑤ Indole	+	—
⑥ pathogenicity For mice and rabbits	+	—

3- Differences between B. anthracis and B. pseudo-anthraxis

Criteria	B. anthracis	B. pseudo-anthraxis
a- species	one species	Many species
b- Morphology		
1- End of bacilli	Truncate (square end)	Convex or round
2- chain formation	Long chain	Single, diplo or short chain
3- shape and position of spore	Oval, central and not bulging	Vary in shape and position with or without bulging.
4- Capsule	Present (polypeptide)	absent
5- Motility	Non-motile	Motile by peritrichous flagella (4-12)
C- Culture characters		
1- growth in broth	Cotton Wool Fluffy deposit	Turbidity or surface pellicle
2- growth on 10 µg/ml penicillin agar	No growth	good growth
3- growth on 0.5 µg/ml penicillin agar	string of pearls (chain of rounded forms)	No change in the shape of organism

Criteria	<i>B. anthracis</i>	<i>B. pseudo-anthraxis</i>
4-growth in gelatin stab	Inverted Fir-tree growth	Fir-tree absent or atypical
5-Haemolysis on sheep RBCs	Weak	often strong
1-Biochemical tests		
1-gelatin Liquefaction	slow	Rapid
2-Lecithinase reaction	Weak +ve	strong +ve
3-MB reduction	Weak	strong
4-Fermentation of salicin	Slow or not at all	Rapid
e-pathogenicity		
1-Lab. animals	pathogenic	Non-pathogenic
2-Man and animals	Anthrax	In man, <i>B. subtilis</i> → Cause severe eye infection. <i>B. cereus</i> → Cause food poisoning
F-susceptibility to gamma phage	susceptible	Insusceptible

4- Differentiation of *Campylobacter* Species:

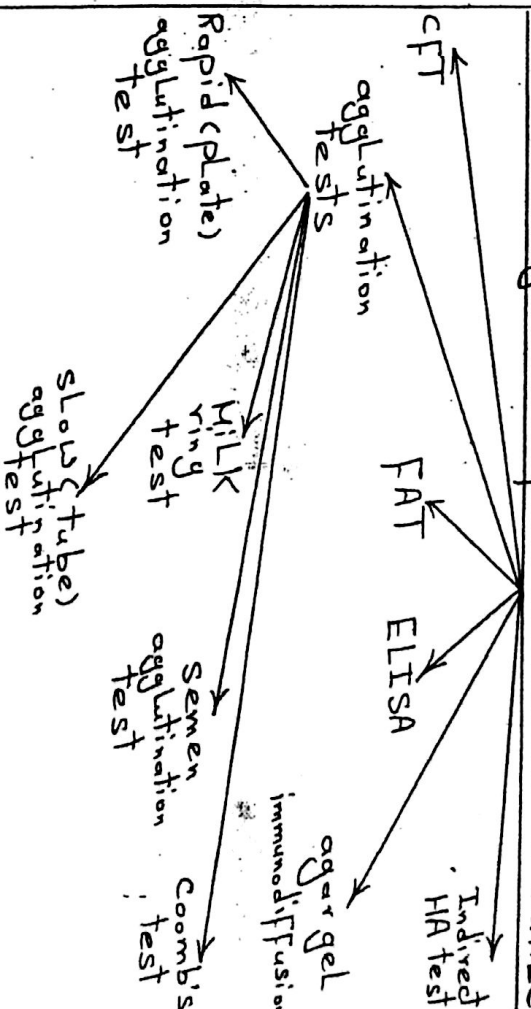
points of differences	C. Fetus		C. jejuni		C. coli		C. sputorum
	ss. Fetus	ss. veneralis					
(a) to differentiate pathogenic and non-pathogenic species							
1-Catalase	+	+	+	+	+	+	+
2-Nitrate reduction	+	+	+	+	+	+	+
3-H ₂ S Production on TSI	—	—	—	—	—	—	+
4-growth in 3.5% NaCl	—	—	—	—	—	—	+
(b) to differentiate pathogenic species							
5-growth in 1% glycine	+	—	+	+	+	+	—
6-growth at 25°C	+	+	—	—	—	—	—
7-growth at 42°C	—	—	+	+	+	+	—
8-Inhibition of growth by:							
• Cephalosporin	+	+	—	—	—	—	—
• Nalidixic acid	—	—	+	+	+	+	—
9-Sodium hippurate hydrolysis	—	—	+	+	+	+	—

5 - Micrococci and staphylococci

points of differences	Micrococci	staphylococci
① oxygen requirement	strict aerobic	facultative anaerobic
② oxidase test	+	-
③ Fermentation of glucose anaerobically	-	+
④ growth on Furazolidon agar.	+	-
⑤ arginine decarboxylase test	-	+
⑥ sensitivity to Lysozyme	+	-
⑦ Teichoic acid in cell wall	-	+
⑧ Lysis by Lysozyme and endopeptidase	-	+
⑨ G+C content of DNA	65 - 75 %	30 - 38 %

③ Comment on :

1 - Serodiagnosis of brucellosis in cattle



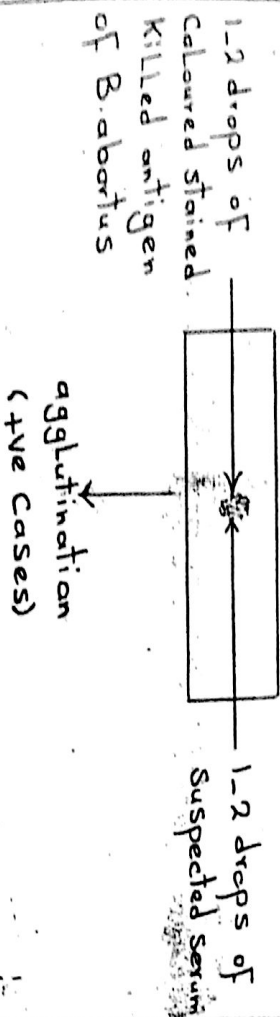
① Complement Fixation test (CFT):

- It is highly specific and Very sensitive depending on IgM and IgG₁
- Complement Fixing antibodies appear in the blood of infected animals shortly before agglutinins.

2 Agglutination tests:-

a. Rapid (plate) agglutination test:

1- Coloured stained Killed antigen of *B. abortus*:
Used for Control of disease and depends on IgM and IgG₁



2- Rose-Bengal plate test:

- antigens → buffered to pH 3.65 - 4
→ stained with Rose-Bengal stain (Red)
- Serum → diluted with acid buffer (pH 3.5) to destroy IgM (non-specific antibodies) leaving IgG (specific antibodies) → so, this test is more accurate.

3- Rivanol test:

depends on precipitation of IgM (mostly produced by adult vaccination) by treating the Serum with Rivanol solution.

b. slow (Tube) agglutination test:

1- ordinary tube agglutination test:

- It is a standard test used for animals positive for plate agglutination test.
 - used routinely in the farms.
 - depends on IgM and IgG₂
 - done by making 2-fold dilutions of Serum
- Result:

	Cattle	Sheep	Human
Suspicious	$\frac{1}{20}$	$\frac{1}{40}$	$\frac{1}{80}$
positive	$\frac{1}{40}$	$\frac{1}{80}$	$\frac{1}{160}$

→ Suspicious cases are re-tested after 10-15 days because agglutinins appear in the blood 10 days post-abortion.

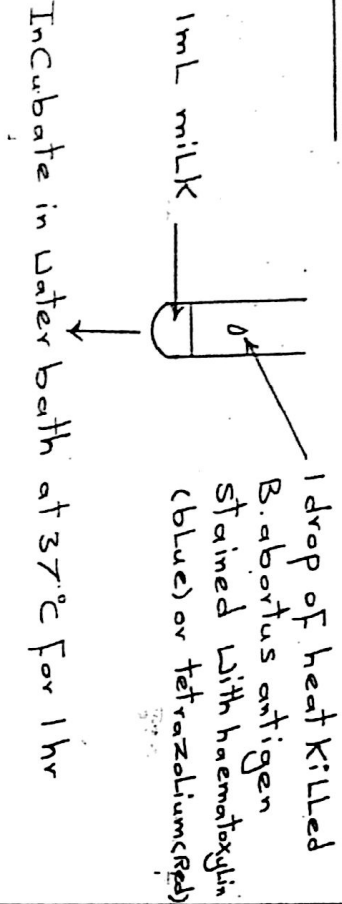
2- Mercaptoethanol test:

In which, IgM antibodies are destroyed (These antibodies are mostly produced by adult vaccination)

c- Milk ring test (Abortus Bang Ring test, ABR)

- depends on the presence of IgM, IgG and IgA
- This test depends on that the agglutinins which are excreted in milk during infection are carried to the surface with the rising fat globules.

procedure:

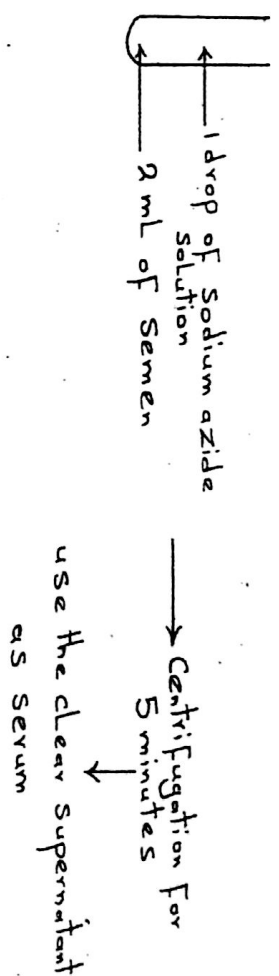


Result:

Cream Layer	Coloured	unColoured	Coloured
Milk	unColoured	Coloured	Coloured
Result	+ve	-ve	suspicious

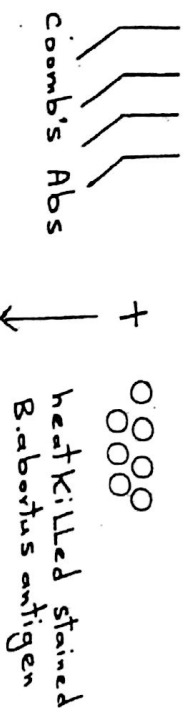
→ Suspicious Cases are retested after 10-15 days

d- Semen agglutination test:

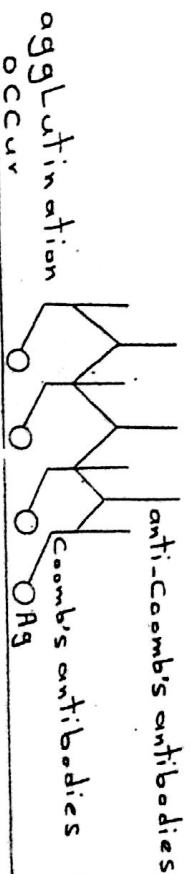


e- Coomb's test:

Very sensitive in Chronic infection → due to the production of Coomb's antibodies (Incomplete antibodies contain one Fab portion only)



So, use anti-Coomb's antibodies (antiglobulins)



2- Isolation of Salmonellae:

1. cultivation on selective enrichment broth:

- Such as
 - Selenite F broth
 - tetrathionate broth
 - Rappaport Vassiliadis broth
- This step is done to enhance the growth of Salmonella and inhibit the growth of other enteric bacteria.

- The inoculated tubes are incubated aerobically at 43 °C or 37 °C.

→ With addition of Na sulphathiazole at 1.25 mg/l
→ The incubation time → Not more than 18-20 hrs to suppress the growth of proteus spp.

2. plating on selective differential solid media:

- Such as
 - MacConKey's agar
 - Salmonella-shigella agar (S-S agar)
 - Desoxycholate citrate agar (DCA)
 - brilliant green agar
 - xylose lysine desoxycholate agar (XLDA)

- The inoculated plates are incubated aerobically at 37 °C for 1-3 days.

- Suspected colonies of Salmonella are picked up and subcultured onto the surface of selective differential solid media to get a pure culture.



Q: How can you obtain a pure culture of an organism suspected to be Salmonellae from intestinal contents (2006)

Identification:

Culture characters:

- on nutrient agar:

Smooth colonies

- on MacConKey's agar:

Salmonellae produce colorless colonies because they are non-lactose fermenter, except S. grizonae (late lactose fermenter, after 3 days)

● on S-S agar: as MacConKey's agar

● on DCA: as MacConKey's agar

● on brilliant green agar:
pink colonies

● on XLD:

Xylose utilization
with acid production
↓
yellow colonies at first

Lysine
decarboxylation
↓
Red colonies

H₂S
production
↓
Black centers

→ Indicator → phenol red indicator

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3 - general characters of Corynebacteria.

(a) Morphology:

- Gram staining reaction → Gram +ve
- acid fastness → Non-acid fast
- shape → pleomorphic (tend to form filaments or thread like forms in old culture)
→ Many spp. have metachromatic granules esp. *C. diptheriae*.
- size → variable in size
- arrangement → Chinese letters or palisade arrangement because during division, the daughter cells remain attached in one side producing L and V arrangement (Chinese letters) or arranged in rods parallel to each other (palisade arrangement).
- Motility → Non-motile
- Capsule → Non-capsulated
- spore → Non-spore

(b) growth requirement:

- O₂ req. → aerobic or facultative anaerobic
- opt. temp. → 37 °C
- opt. pH → Neutral
- Incubation time → 1-3 days
- growth on ordinary media → -ve
- growth on MacConKey's agar → -ve
- tolerance to NaCl → -ve

C Biochemical activities:

- catalase —————→ +ve
- oxidase —————→ -ve

4) How Can you:

- 1- classify Mycobacteria
- 2- classify clostridia
- 3- Vaccinate against brucellosis in cattle

Classification of mycobacteria:

I The classic species (Typical T.B.):

They have been recognized for many years as causes of diseases in man and animals.

• They include:

- 1- M. bovis (Bovine tubercle bacillus)
- 2- M. avium (avian tubercle bacillus)
- 3- M. tuberculosis (Human tubercle bacillus)
- 4- M. paratuberculosis (John's bacillus)
- 5- M. leprae (Hansen's bacillus)

II Atypical (unclassified or anonymous) mycobacteria (Runyon's groups):

classified into 4 groups acc. to rate of growth and pigment production:

a- slow growers:

group I (photochromogenic):

- produce yellow pigmented colonies only after exposure to light.
- slow growing (require 7 days or ↑ for visible growth to be seen).
- e.g. M. Kansasii and M. marinum.

group II (scotochromogenic):

- produce yellow or orange pigment either in the presence or absence of light.
- Slow growing
- e.g. *M. goodii*, *M. xenopi* and *M. scrofulaceum*

group III (Non-photochromogenic):

- produce no or slight pigment with exposure to light.
- slow growing
- e.g. *M. ulcerans*

b. Rapid growers:

group IV (variable pigmentation):

- give variable pigmentation
- grow rapidly (visible growth seen in less than 7 days)
- e.g. *M. fortuitum*, *M. phlei* and *M. smegmatis*

III Saprophytic mycobacteria:

- Non-pathogenic mycobacteria
- widely distributed in nature (soil, grass, water)
- resemble T.B but differ in that they grow well at room temp. and on nutrient agar.
- e.g. *M. phlei* and *M. smegmatis*

2 - classification of clostridia:

① acc. to biochemical activities on proteins and CHO in cooked meat medium:

1- proteolytic group:

decompose protein and turns meat particles into black colour with foul odour.
e.g. *C. histolyticum*

2- Saccharolytic (gas gangrene) group:

Ferment CHO in meat → pink colour with production of large amount of gases (stormy fermentation) e.g.
→ *C. perfringens*
→ *C. septicum*
→ *C. novyi*
→ *C. chauvoei*

3- proteolytic and saccharolytic group:

e.g. *C. botulinum*

4- Non-proteolytic and non-saccharolytic group:

e.g. *C. tetani*

② acc. to pathogenicity:

a - Saprophytic clostridia:

- Commonly Found in soil, sewage and water in sporulated form.
- However, pathogenic clostridia (such as *C. tetani*, *C. septicum*, *C. chauvoei* and *C. novyi*) Found in soil as saprophytes.

b - pathogenic clostridia:

- Normal inhabitant in the intestinal tract of man and animals and produce the disease under certain conditions.
- divided into 2 groups acc. to mechanism of disease production:

Invasive (gas gangrene)

group

invade and multiply in internal organs with production of large amount of less potent toxins such as:

- C. perfringens*
- C. septicum*
- C. chauvoei*
- C. novyi*

Non-invasive (highly toxic)

group

have no power to invade living tissues.

Their pathogenicity depends on production of highly powerful neurotoxins either in:

- ① Localized infected deep wound (*C. tetani*)
- ② Contaminated Canned or salted Fish (*C. botulinum*)

③ acc. to position of spore and gelatin

Liquefaction:

Subterminal spores with gelatin hydrolysis

group

- C. botulinum*
- C. perfringens*
- C. chauvoei*
- C. septicum*
- C. novyi*

Terminal spores with gelatin hydrolysis

C. tetani

Terminal spores without gelatin hydrolysis

include saprophytic clostridia e.g. *C. tertium*

Subterminal spores without gelatin hydrolysis

include saprophytic clostridia e.g. *C. butyricum*

② acc. to pathogenicity:

a - Saprophytic clostridia:

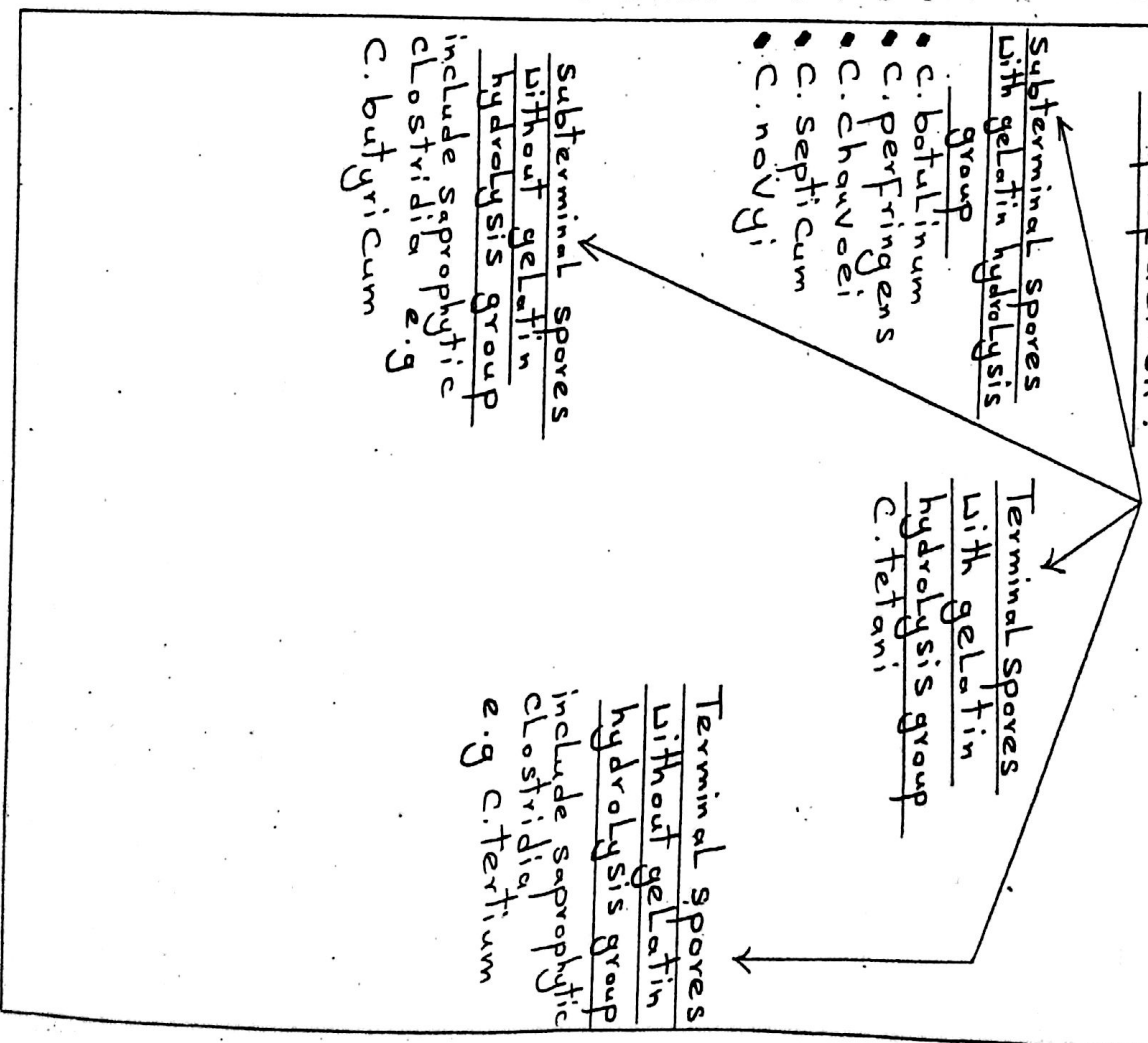
- Commonly Found in soil, sewage and water in sporulated form.
- However, pathogenic clostridia (such as *C. tetani*, *C. septicum*, *C. chauvoei* and *C. novyi*) found in soil as saprophytes.

b - pathogenic clostridia:

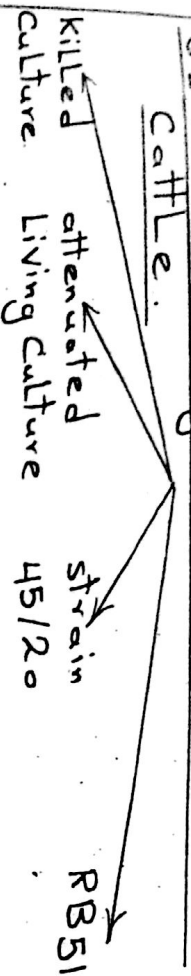
- Normal inhabitant in the intestinal tract of man and animals and produce the disease under certain conditions.
- divided into 2 groups acc. to mechanism of disease production:

Invasive (gas gangrene) group	Non-invasive (highly toxic) group
<ul style="list-style-type: none"> invade and multiply in internal organs with production of large amount of less potent toxins such as: <i>C. perfringens</i> <i>C. septicum</i> <i>C. chauvoei</i> <i>C. novyi</i> 	<ul style="list-style-type: none"> have no power to invade living tissues. Their pathogenicity depends on production of highly powerful neurotoxins either in: ① Localized infected deep wound (<i>C. tetani</i>) ② Contaminated canned or salted fish (<i>C. botulinum</i>)

③ acc. to position of spore and gelatin liquefaction:



3- Vaccination against brucellosis in cattle.



1- Killed culture (Bacterin):

- Killed culture of *B. abortus* either by heat or chemicals with oil adjuvant.
- It is not harmful and animals are vaccinated twice by it.
- produce neither solid nor lasting immunity because it produces humeral immunity but not cell-mediated immunity (which is protective).
- produce Complement Fixing antibodies.

2- attenuated Living culture:

→ *B. abortus* strain 19 (Book strain) → used for immunization of cattle at age of 6-8 months old → S/C (single injection)

- animals < 5 months old should not be

vaccinated:

- 1- to avoid the interference with maternal immunity.
- 2- cells and tissues of the immune system will not be so highly developed to form a level of Abs sufficient to produce good protection.

disadvantages:

- ① adult animals develop permanent agglutinins which interfere with agglutination test results → so, it is difficult to differentiate between the diseased one and the vaccinated one.
- ② may cause severe postvaccination reaction → abortion in pregnant cattle.
- ③ contraindicated in vaccination of ♂ at any age
 - because it produce permanent agglutinin and so can't differentiate between the infected male and vaccinated one.
 - may cause infertility in some ♂ calves

3- strain 45/20 :

- It is rough strain of *B. abortus*
- The strain No. 45 is subcultured 20 times.
- disadv.:
- Live rough strain of *B. abortus* 45/20 is virulent in vivo.

4- RB51

⑤ What are the Virulence Factors of?

- 1- streptococci
- 2- Members of *F. enterobacteriaceae*

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Virulence Factors of streptococci

Products
(Exotoxins and Enzymes)

Extracellular structures
in bacterial cell

① Products (Exotoxins and enzymes):

① Haemolysin (streptolysins):

streptolysin "O"	streptolysin "S"
oxygen - labile	oxygen - stable
haemolysis of anaerobically incubated sheep blood agar.	β-haemolysis of aerobically incubated blood agar
produced by streptococci of groups A, C and G but not by members of other groups.	produced by streptococci of groups A, C and G and also by members of other groups esp. pyogenic ones.

It is protein in nature (i.e. antigenic) → So, ASO (anti-streptolysin "O") is formed in the serum of patients suffering from infection with *S. pyogenes* or other streptococci producing streptolysin "O".

Not antigenic

② streptokinase (Fibrinolysin):

- produced by streptococci groups A, C and G
- cause digestion (lysis) of Fibrin → through conversion of plasminogen into plasmin which lyse the Fibrin.

③ Erythrogenic (pyrogenic) toxins:

- produced by group A streptococci
- They are 3 types (A, B and C) which act as Superantigens.
- responsible for the characteristic skin rash of scarlet fever.
- antigenic

④ Hyaluronidase (spreading factor):

- produced by groups A, B and C streptococci
- aid in the spreading of streptococci → as it splits hyaluronic acid (continuity substance between cells and tissue spaces)

⑤ other extracellular substances:

- NADase
- DNase
- Protease
- serum opacity factor
- C5a peptidase → It degrades C5a which is the most important chemotactic peptide → So, inhibit phagocytosis.

II Extracellular structures in bacterial cell:

1- Capsule:

- Found in S. pyogenes, S. pneumoniae, S. agalactiae and S. equi subsp. equi.
- It is antiphagocytic.

2- M-protein:

- antiphagocytic (S. pyogenes and S. porcinus)
- adhesin (S. equi subsp. equi)
- cytotoxic to neutrophils.
- bind factor H in serum (regulatory protein for the alternative complement pathway) → thus, C3b that binds to the cell surface in the region of M-protein is degraded by factor H.
- Part of it is found in fimbriae → binding Fibrinogen from serum → blocking the binding of complement to the underlying peptidoglycan → inhibit phagocytosis.
- surface M-proteins contain antigenic epitopes → attach to heart muscle → leading to autoimmune rheumatic carditis (Rheumatic fever).
- It is strongly immunogenic and the formed antibodies are protective → but, they are more than 80 antigenic types → therefore, the development of vaccine is not feasible.
- There are 80 serotypes of S. pyogenes acc. to M-protein (Griffith classification)

3- Lipoteichoic acid (LTA):

- act as adhesin.
- present in the Fimbriae

4- F-protein (Fibronectin-binding protein):

- act as adhesin → Causing bacterial attachment to epithelial cells of pharynx and skin.
- Fibronectin → a matrix protein on eukaryotic cell.

2-

Virulence Factors of Enterobacteriaceae

<u>Factors related to bacterial cell</u>	<u>Factors related to bacterial products</u>
<ul style="list-style-type: none"> → Endotoxins → Capsule → Fimbriae → Invasiveness 	<ul style="list-style-type: none"> → Exotoxins → Haemolysin → Siderophores (chelating agents)

A) Factors related to bacterial cell

① Endotoxins:

- They are LPS (represent the outer layer of cell wall)
 - the main endotoxic principle is Lipid_A → which interfere with the Complement Components.
 - they are released when bacteria die and lyse (either inside or outside phagocytic cells)
 - you have to take in consideration death of bacterial cells outside phagocytic cells like Complement-mediated lysis or antibiotic-mediated lysis
- Cause
- 1- Fever
 - 2- Leukopenia followed by Leukocytosis
 - 3- hyperglycemia then hypoglycemia and lethal shock after a latent period.

capsule:

- It is antiphagocytic substance where its surface is hydrophilic which repel hydrophobic surface of phagocytes
- Interfere with binding of Abs to bacteria.
- Poor activator of complement → protect cell wall of bacteria from damaging effects of complement.

③ Fimbriae (pili, adhesins):

- allow adhesion of bacteria to host cells with subsequent colonization → such as those in E. coli allow its attachment to glycoproteins found on the surface of epithelial cells of jejunum and ileum followed by colonization → so produce its pathogenicity.

④ Invasiveness:

- ability of M.O to attack host cells.
- Invasive strains such as Salmonella typhi and S. dublin → are able to attack the host cells causing their damage → bloody diarrhea.
- It is plasmid (genetically) - controlled.

② Factors related to bacterial products

① Exotoxins:

<u>Enterotoxins</u>		<u>Neurotoxins</u> <u>(Cytotoxins, shiga-like toxins)</u>
2 types acc. to the effect of heat:		
<u>Labile type</u> <u>(LT)</u>	<u>stable type</u> <u>(ST)</u>	
destroyed by heating at 60°C/30 min.	destroyed by heating at 121°C/15 min.	→ destroy vascular endothelial cells causing bloody diarrhea. → they inhibit protein synthesis in host cells by interaction with 60S ribosomal subunit.

② Haemolysin:

- α-haemolysin causes damage to the host cell membranes.

3) Siderophores (chelating agents):

- They are low molecular mass (HM) Iron binding compounds produced by some bacteria → which enable them to utilize the iron required for their growth.
- They aid the bacteria in the competition with host for iron (which is an absolute requirement for the growth of most bacteria).
- Bacteria produce Iron-regulated outer membrane (OM) proteins (IROMPs) → responsible for the internalization of siderophore-iron compounds into bacterial cell.

6) Write on:

1. Ascoli test:

aim:

Thermo-precipitation test used for diagnosis of anthrax in dead animals (old dry skin, hides or other tissues)

Principle:

detect the presence of heat-stable polysaccharide antigen of B. anthracis using specific anthrax antiserum.

procedure:

1. extraction of antigen:

Little Saline Homogenization → boiling for 15 min. or 0.5% acetic acid (5-10 ml)
2-3 g of the suspected tissue

Filtration
↓
clear fluid

2. specific anthrax antiserum:

Prepared in large animal (horse) with virulent culture of B. anthracis.

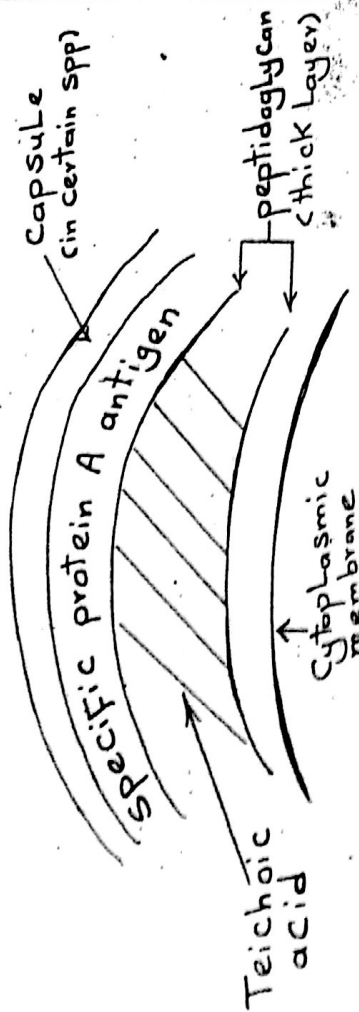
3. Technique:

the Ag extract is introduced into capillary tube and overlaid with anthrax-antiserum (equal amount, 1ml for each)

Result:

+ve reaction → Formation of a thick white ring at the junction of 2 liquids within 15 min. at room temp.

2-CELL WALL Composition of staphylococci:



● It is composed of 3 Layers:

① Capsule:

- present in certain spp (*S. aureus*)
- Consists of polysaccharide (slime layer)
- microcapsule → seen by EM

② peptidoglycan:

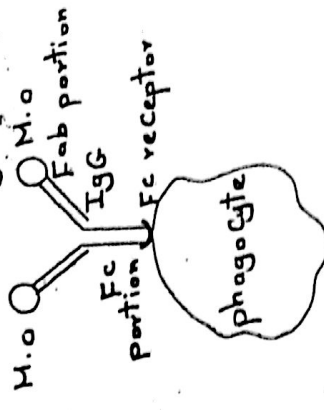
- Contains L-Lysine and Teichoic acids
- Teichoic acids may be → Ribitol (*S. aureus*, *S. saprophyticus*)
- glycerol (*S. epidermidis*)

Functions of teichoic acids:

- 1- protection against action of Lysozyme.
- 2- site of bacteriophage adhesion.

③ Specific protein A antigen (SPA):

- It is the antigenic part of cell wall
- Constituting 40-60% of cell wall
- $\frac{2}{3}$ SPA in cell wall
- $\frac{1}{3}$ SPA in culture filtrate (extracellular SPA)
- It binds with FC portion of immunoglobulins (esp. IgG) → this is called pseudoimmune reaction where a true immune reaction is the binding with Fab portion of immunoglobulin



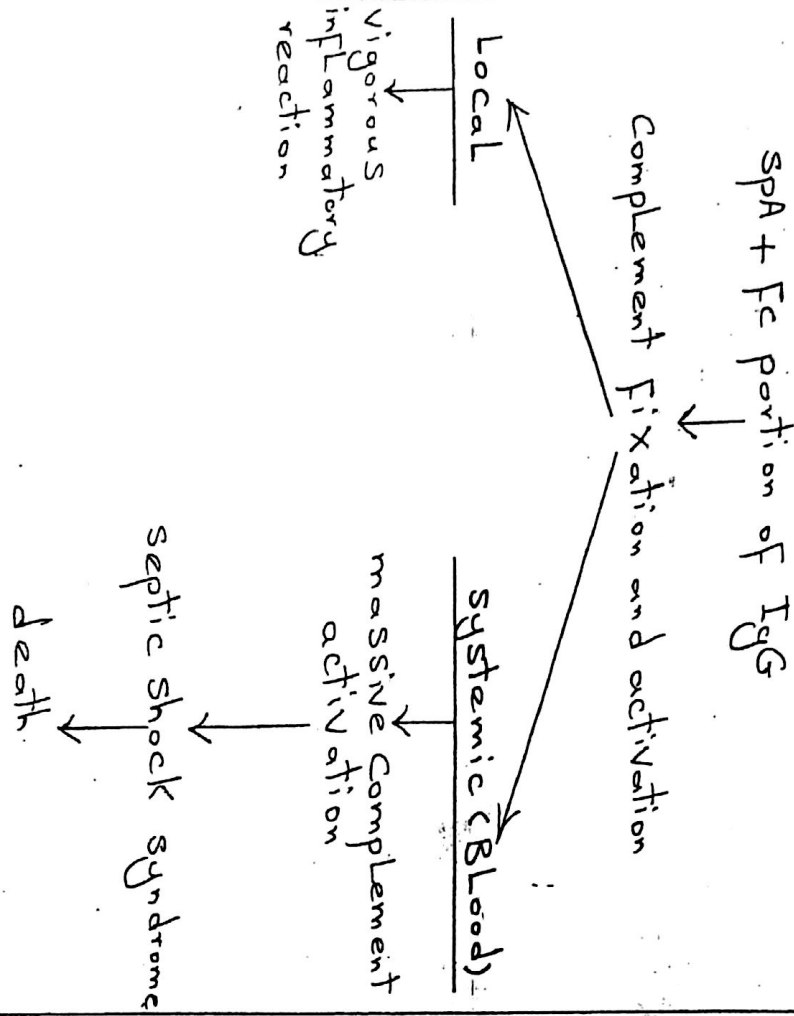
"No phagocytosis"

True immune reaction

→ This binding hinders FC-mediated opsonization (No phagocytosis), however Complement activated by protein A-bound immunoglobulin can contribute to vigorous inflammatory reaction.

- When *S. aureus* reach blood stream both protein A-bound immunoglobulin and

exposed peptidoglycan can induce massive complement activation contributing to the pathogenesis of septic shock syndrome → death



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3- Types of pathogenic E. coli

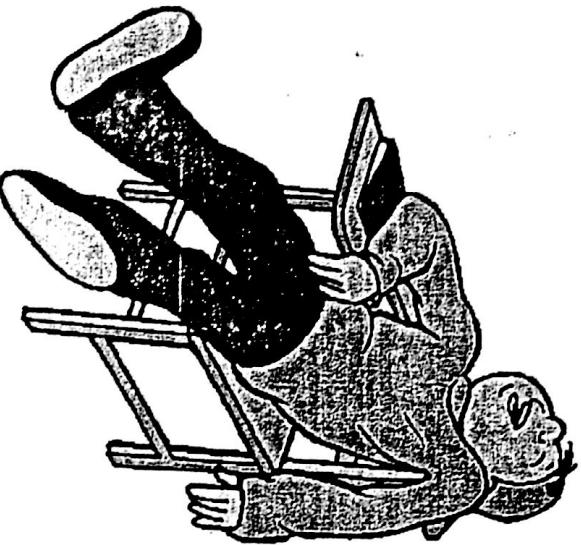
→ there are 5 types acc. to mechanism of gastroenteritis

Entero-toxigenic E. coli (ETEC)	Entero-invasive E. coli (EIEC)	Entero-pathogenic E. coli (EPEC)	Entero-aggregative E. coli (EA _{agg} EC)	Entero-haemorrhagic E. coli (EHEC)
→ produce <u>enterotoxins</u> (which may be LT or ST) → <u>genetically-controlled</u> (plasmid-controlled) → F-antigens are attached to the surface of intestinal epithelial cells → for toxin production. → Cause gastroenteritis which is ch' by: ① vomiting ② secretory diarrhea (traveler's diarrhea) → similar to that caused by <i>Vibrio cholerae</i> . → <u>Cause Food poisoning in man.</u>	→ <u>Invade</u> the intestinal epithelial cells. → plasmid-controlled → Cause <u>bloody diarrhea</u> (dysentery-like disease) similar to that caused by <i>shigella</i> species	→ Contain <u>adherence factor plasmid</u> → It <u>adheres</u> to the intestinal epithelial cells → this adhesion is followed by inflammation. → Cause <u>diarrhea in lambs and children.</u>	stacked brick like aggregative pattern of adherence is formed due to the presence of <u>aggregative adherence plasmid</u> .	→ produce <u>Cytotoxins</u> (shiga-like toxin) which damage the vascular endothelial cells in the intestinal tract causing <u>bloody diarrhea</u> in humans similar to <i>shigella</i> dysenteriae → so, it is called <u>haemorrhagic colitis in man.</u>

Systematic Bacteriology

[Exams Guide]

امتحان ٢٠١٢



Bacteriology 2012

Answer the following questions :

1) why are the following sentences incorrect (x) ?

1. Strangles in equines is caused by *Burkholderia mallei*.
2. *Brucella* species are acid fast organisms.
3. *Mycoplasmas* are sensitive to penicillins.
4. Vaccination of pregnant ewes against lamb dysentery is active for lambs.
5. Tetanus must occur by oral infection through ingestion of food.
6. Spores of *S. pyogenes* are found subterminal.

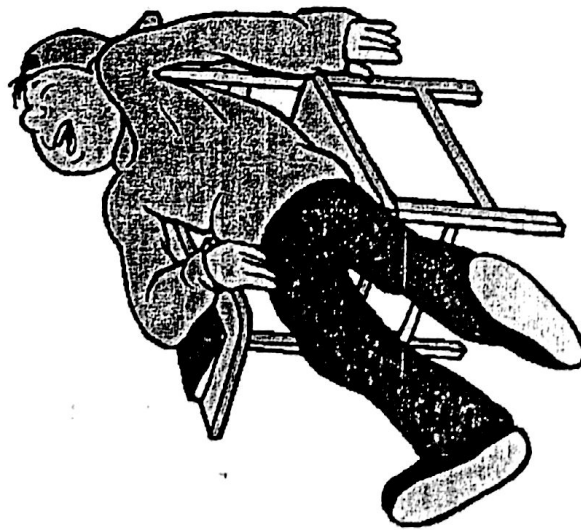
① strangles in equines is caused by *streptococcus equi* subsp. *equi* or glanders in equines is caused by *Burkholderia mallei*

② *Brucella* spp. are non-acid fast organisms because they don't resist decolorization by strong acid (conc. H_2SO_4) and don't stain red by Ziehl Neelsen's stain. but, they are weak acid fast organisms because they resist decolorization by weak acids (such as 0.5% acetic acid) and stain red by modified Ziehl Neelsen's stain.

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3) Mycoplasmas are resistant to penicillins because they lack cell wall and don't synthesize peptidoglycan.

4) Vaccination of pregnant ewes against Lamb dysentery is active for mothers and passive for Lambs → as pregnant ewes receive 2 doses of formalized whole culture of *C. parvum* type B (1st dose at 5 wks before parturition and the 2nd dose at 2 wks after 1st dose) → after parturition, the Lamb must receive the Colostrum from the immunized dams because it contains high conc. of B and E antitoxins (natural passive immunity).

5) Tetanus must occur by deep wound infection

6) *S. pyogenes* does not form spore

2) how can you differentiate.....?

1. *Listeria* from *Corynebacteria*.
2. *Pseudomonas* from each other.

1- *Listeria* from *Corynebacteria*

	<i>Listeria</i>	<i>Corynebacteria</i>
• Morphology		
• Gram staining reaction	Gram +ve	Gram +ve
• acid fastness	Non acid fast	Non-acid fast
• shape	Coccobacilli	pleomorphic (tendrils, form filaments or thread-like forms in old culture)
		• Many spp have metachromatic granules esp. <i>C. diphtheriae</i>
• size	Small to medium	Variable in size
• arrangement	uncharacteristic	Chinese letters or palisade arrangement → because during division, the daughter cell remain attached in one side producing L and V arrangement (Chinese letters) or arranged in rows parallel to each other (palisade arrangement)
• Motility	motile by a few (1-5 peritrichous flagella) (tumbling motility)	Non - motile
• Capsule	Non - Capsulated	→
• spore	Non - spore	→

	Listeria	Corynebacterium
⑧ growth requirement:		
1- O_2 requirement	Facultative anaerobes but growth is enhanced in the presence of 10% CO_2 (Capnophilic)	aerobic or facultative anaerobic
2-opt. temp.	3-45°C (opt. temp. 37°C) (cold enrichment)	37°C
3-opt. pH	5.6 - 9.6	Neutral
4-Incubation time	1-3 days	1-3 days
5-growth on ordinary media	grows on nutrient agar	-ve
6-growth on MacConkey's agar	-ve	-ve
7-Tolerance to NaCl	tolerate 10% NaCl (Halophilic organism)	-ve
⑨ pigment production	-ve	endopigment producer (yellowish white)
⑩ Biochemical activities	Catalase +ve oxidase -ve hydrolyze aesculin	Catalase +ve oxidase -ve

30

2-

Differences among pseudomonads and Burkholderiales

points of differences	B. mallei	B. pseudomallei	P. aeruginosa	P. fluorescens
① Motility	Non-motile	Motile by mono- or lophotrichous flagella	Motile by monotrichous flagella	Motile by lophotrichous flagella
② gelatin Liquefaction	-	+	+	+
③ growth at: ♦ 5°C (Low temp) ♦ 42°C (high temp)	- - -	- + +	- + +	+ - -
④ pigment	brown	brown	pyocyanin and Fluorescin (bluish green)	Fluorescin (yellowish green)!
⑤ growth on MacConkey's agar	-	+	+	+
⑥ oxidation of Lactose	-	+	-	-
⑦ pathogenicity	glanders (equines)	Melioidosis (Sheep)	Variety of pathological conditions	pseudomonas disease in Fish
⑧ diagnosis in Living animal	Mallein Test	Melioidin Test	-	-
⑨ arginine dihydrolase Test	+	+	+	+
⑩ pH 6	+	+	-	-

3) choose between brackets to complete the following sentences?

1. Tet toxin is produced only by C. perfringens type E.
2. Milk ring test is used for diagnosis of (salmonellae, listeriae, brucellae, corynebacteria, mycobacteria) in raw milk.
3. Neurotoxin that produced by C. tetani binds (reversibly, irreversibly) with acetyl choline to the gangliosides of nerve cells, so, antitoxin is (highly, mostly, not, less) effective.
4. Generally, pathogenic clostridia give (positive, negative, suspected) results with gelatin liquefaction test.
5. M-protein is present in the cell wall of (S. aureus, S. pyogenes, E. coli, A. pyogenes).
6. Different species of pseudomonads are (less active, active, of no value) in their results for carbohydrate fermentation.
7. Digitonin test is an important test for (salmonellae, listeriae, mycoplasmas, brucellae, spirochaetes).
8. is a commensally potential pathogen (Salmonella Enteritidis, Brucella abortus, Mycobacterium tuberculosis, pasteurella multocida).
9. ETEC differs from EIEC in (motility, capsule, enterotoxins production, microscopic appearance.)

4) put the mark (✓) or (x) in front of each of the following sentences?

1. There is a strong relationship between motility of salmonellae and their pathogenicity. ✓
2. Tetanolysin is only produced by all toxigenic strains of C. perfringens. X
3. Symptomatic anthrax is a disease infecting sheep and caused by B. anthracis type A through ingestion of its spores. X
4. Bacillary haemoglobinuria in cattle is a disease caused by C. novyi type D. ✓
5. Chickens and pigeons are susceptible to infection by B. abortus. X
6. Fried egg microcolonies is a character of mycoplasmas. ✓

5) Mention the cause(s) and explain your answer:

1. Food poisoning in humans arises from eating salted fish.
2. Septic shock syndrome.

① Contaminated salted fish may contain highly powerful neurotoxins of Cl. botulinum (which are acid-stable so, not-affected by gastric juice) → absorbed through digestive mucosa → symptoms appear after 36-96 hrs.

② septic shock syndrome

occurs due to massive complement activation induced by SPA-bound immunoglobulin and exposed peptidoglycan when S. aureus reach blood stream.

• SPA → is the antigenic part of cell wall of S. aureus, and it binds with Fc portion

of immunoglobulins (specially IgG) →
this is called pseudo immune reaction.

6) Give an idea on:

1. Role of rodents in the transmission of some pathogenic bacteria.

Infected rodents (wild rat and mice) excrete Salmonellae in their feces contaminating both human and animal feed as well as general environment → Cause Food poisoning (Food infection)

2. Lipids in the cell wall of mycobacteria and their importance.

a) Mycolic acids:

They are strong hydrophobic molecules that form a lipid shell around the organism.

- 1- responsible for acid fastness (the property of retaining Carbol Fuchsin after the application of decolorizing acid and alcohol)
- 2- a significant determinant of virulence.
- 3- prevent attack of mycobacteria by cationic proteins, lysozyme and oxygen radicals in the phagocytic granule.
- 4- protect extracellular mycobacteria from complement deposition in serum.

b) Mycosides:

- responsible for control of cellular permeability → resistance to water soluble enzymes, antibiotics and disinfectants.

• Wax D:

- Amycoside that enhance immune response.
- induce delayed hypersensitivity

c) glycolipids:

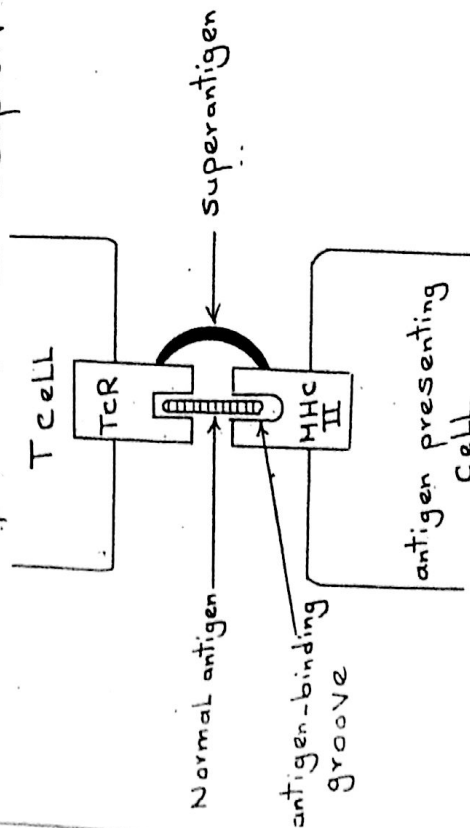
- result in toxicity and granulomatous response.
- enhance survival of phagocytosed mycobacteria.
- Cord factor → a glycolipid responsible for characteristic colonial growth (Long palisade like growth resembling serpentine cord).
- Sulfatides → inhibit phagolysosome formation permitting T.B to survive intracytoplasmically after being ingested by macrophages.

3. Staphylococcal superantigens. (Toxic shock Syndrome)

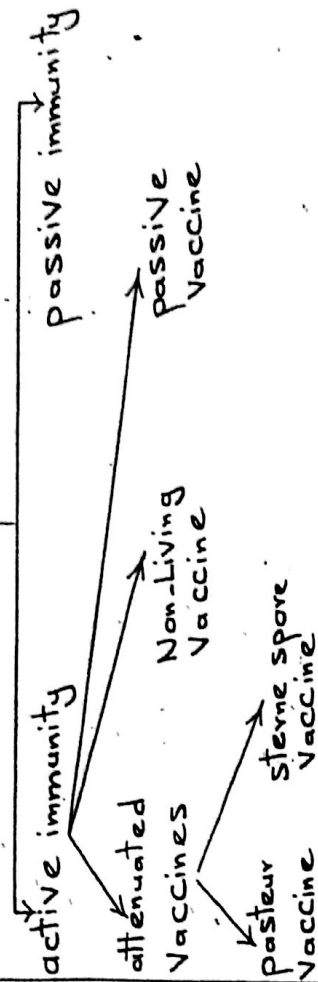
Enterotoxins

TsST-1

- Enterotoxins and TsST-1 are superantigens that may cause toxic shock Syndrome.
- Superantigens stimulate T-cells non-specifically then cytokines are released in large amounts causing symptoms of toxic shock Syndrome.
- Superantigen bind directly to MHC II on APC outside the conventional antigen binding groove. this complex recognize only the V β element of the T-cell receptor.



4. Vaccination against B. anthracis.



I active Immunity:

a-attenuated Vaccines:

1-pasteur Vaccines:

Pasteur I
Live culture attenuated by attenuated for shorter time
Cultivation at 42-43°C (10 days)
For a long time (15-20d)

Pasteur II

Not used nowadays (due to Live H.O)

2-avirulent spore vaccine (Sterne spore vaccine)

avirulent non-capsulated spore forming anthrax bacilli → 10 million spores/ml

Preparation:

- 1- Avirulent non-capsulated spore forming anthrax bacilli are washed from agar plates.
- 2- The suspension is glycerinated to kill the vegetative bacilli.
- 3- dilution of suspension with saponin ($1/8$) in saline to give 10 million spores/mL.
- Saponin \rightarrow induce necrotic reaction which limit the spread of injected M.O

dose:

- 1- Large animals \rightarrow 1ml S/C (10 million spore)
- 2- Small animals (sheep-goat-pig) \rightarrow 0.5 ml (5 million spore)

b. Non-Living Vaccine:

protective Ag. of *B. anthracis* which occurs in sterile oedema fluid and extracts of infected tissue is immunogenic.
 \rightarrow used to protect workers at high risk.

C-passive Vaccine:

- developed by Crystal, et al. (Cornell university)
- passive immunotherapy with adenovirus-based vectors expressing anti-PA (protective antigen) antibody.

- give immunity to anthrax within 24 hours of vaccination.
- Could be given alone or in combination with
 - 1- active vaccines
 - 2- or with antibiotics (in case of anthrax)

II passive Immunity:

using anti-anthrax serum prepared in horses, mules and donkeys.

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5. How can you obtain a pure culture of an organism suspected to be salmonellae from intestinal contents.

1. Cultivation on selective enrichment broth:

- Such as
 - Selenite F broth
 - tetrathionate broth
 - Rappaport Vassiliadis broth

• This step is done to enhance the growth of Salmonella and inhibit the growth of other enteric bacteria.

• The inoculated tubes are incubated aerobically at 43°C or 37°C.

→ With addition of Na Sulphathiazole at

1.25 mg/L

→ The incubation time → Not more than 18-20 hrs to suppress the growth of proteus spp.

2. Plating on selective differential solid media:

- Such as
 - MacConkey's agar
 - Salmonella-shigella agar (S-S agar)
 - Desoxycholate Citrate agar (DCA)
 - brilliant green agar
 - xylitol lysine desoxycholate agar (XLDA)

• The inoculated plates are incubated aerobically at 37°C for 1-3 days.

• Suspected colonies of Salmonella are picked up and subcultured onto the surface of selective differential solid media to get a pure culture.



Q: How can you obtain a pure culture of an organism suspected to be salmonellae from intestinal contents (2006)?

Identification:

Culture characters:

• on nutrient agar:
Smooth colonies

• on MacConkey's agar:

Salmonellae produce colorless colonies because they are non-lactose fermenter except S. arizonae (late lactose fermenter, after 3 days)

• on S-S agar: as MacConkey's agar

• on DCA: as MacConkey's agar

• on brilliant green agar:
pink colonies

• on XLD:

xylose utilization
with acid production
↓
yellow colonies
at first

↓
lysine
decarboxylation
↓
red colonies

↓
H₂S
production
↓
black
centers

→ Indicator → phenol red indicator

7) Discuss:

1. Laboratory diagnoses of spirochaetes. ail

2. Streptococcal toxins.

① Haemolysin (streptolysins):

streptolysin "O"	streptolysin "S"
oxygen - Labile	oxygen - stable
haemolysis of anaerobically incubated sheep blood agar produced by streptococci of groups A, C and G but not by members of other groups	B-haemolysis of aerobically incubated blood agar produced by streptococci of groups A, C and G and also by members of other groups esp. pyogenic ones.
It is protein in nature i.e. antigenic → So, ASO (anti-streptolysin "O") is formed in the serum of patients suffering from infection with S. pyogenes or other streptococci producing streptolysin "O"	Not antigenic

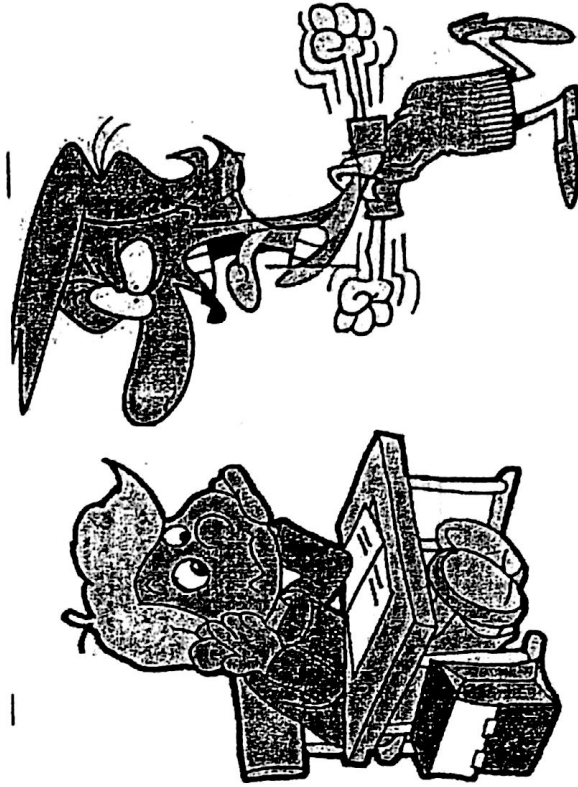
② Erythrogenic (pyrogenic) toxin:

- produced by group A streptococci
- They are 3 types (A, B and C) which act as superantigens.
- responsible for the characteristic skin rash of scarlet fever.
- antigenic

Systematic Bacteriology

(Exams Guide)

امتحان ٢٠١١



different serotypes of *P. multocida* and their pathogenicity in different hosts

Robert's	K-antigen (Carter)	O-antigen (Namioka and Burner)	Host	disease
II	A	5	Fowl	Fowl cholera
II	A	8	Fowl	
II	A	9	Fowl	
I	B	6	Bovine	Haemorrhagic septicaemia
I	E	6	Bovine	
IV	D	1	sheep and swine	Pasteurellosis (shipping fever)
IV	D	4		
III	C	Not available	Not available	Not available

Bacteriology 2011

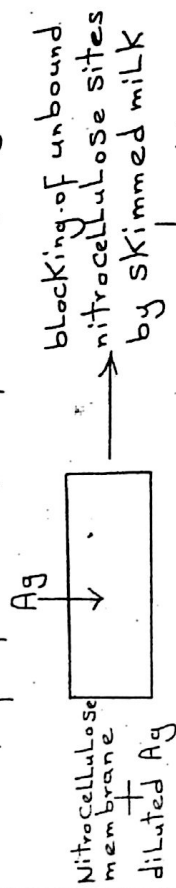
1- Comment on (answer only 3 points)

1- Serodiagnosis of *B. mallei* infection in equidae.

① CFT

② Dot ELISA:

Time to perform → is from 2-7 days



add anti-horse washing 3 times add horse serum antibody (IgG) ← by PBS-Tween 20 under test

Washing →

add conjugate (Horse radish peroxidase)

Washing → add substrate

→ +ve reaction (Brown dot)

③ agglutination test:

Not +ve for 7-10 days and a high background titer in normal sera (1:320 to 1:640) makes interpretation difficult.

• Titer 1:1000 → indicate infection

2- Serodiagnosis of brucellosis in cattle.

3- General characters of mycoplasmas.

① Gram -ve bacteria

② stain poorly with Gram's stain but they produce better results with Giemsa and Romanowsky stains.

③ Intracellular bacteria

④ They are devoid of cell wall peptidoglycan → So, they have not constant shape (pleomorphic).

⑤ The cytoplasm is surrounded by plasma membrane which is a triple-layered and contain sterol (unit-membrane).

⑥ They are the smallest bacteria (0.1-0.3 μm in diameter).

⑦ Non-motile

⑧ growth requirements:

1- O₂ requirement:

• aerobic or facultative anaerobic

• Some of them require 5% CO₂ to grow (capnophilic)

• others are strict anaerobic (called anaerophiles)

2- opt. temp for growth → 35 - 37 °C

3- opt. pH → 7.2 - 7.8

4- They need:

• high conc. of Serum (10-15%), cholesterol or NAD Factor to grow.

انظر لصفحة 2013

• Inhibitors e.g. → Penicillin (kill G⁺ve bacteria)
→ Thalium acetate (kill G⁻ve)
→ amphotericin B (kill fungi)

This is because mycoplasmas lack cell wall
→ so, they are resistant to penicillin that
used for the inhibition of contaminants.

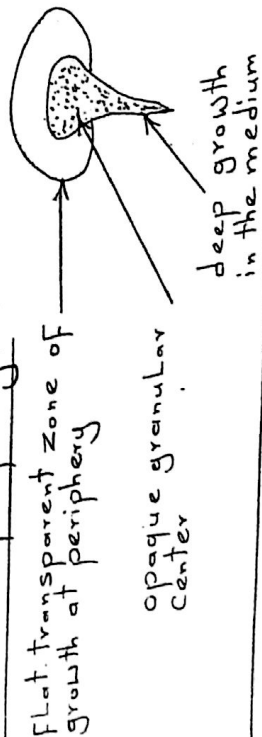
5- Media used:

- Mycoplasma agar and broth
- PPLO media

→ produce Fried egg micro-colonies

• They are the smallest colonies (0.1-0.6 mm in diameter) → visible under dissecting or stereomicroscope or low power of ordinary microscope or hand lens.

• The micro-colonies are ch' by → an opaque granular center with a deep growth in the medium and a flat transparent zone of growth at periphery.



4- General characters of corynebacteria.

2) How can you make? (Answer only 3 points)

1- Classification of mycobacteria.

2- Classification of clostridia.

3- Vaccination against mycobacterium tuberculosis infection.

BCG vaccine (Bacille Calmette Guérin)

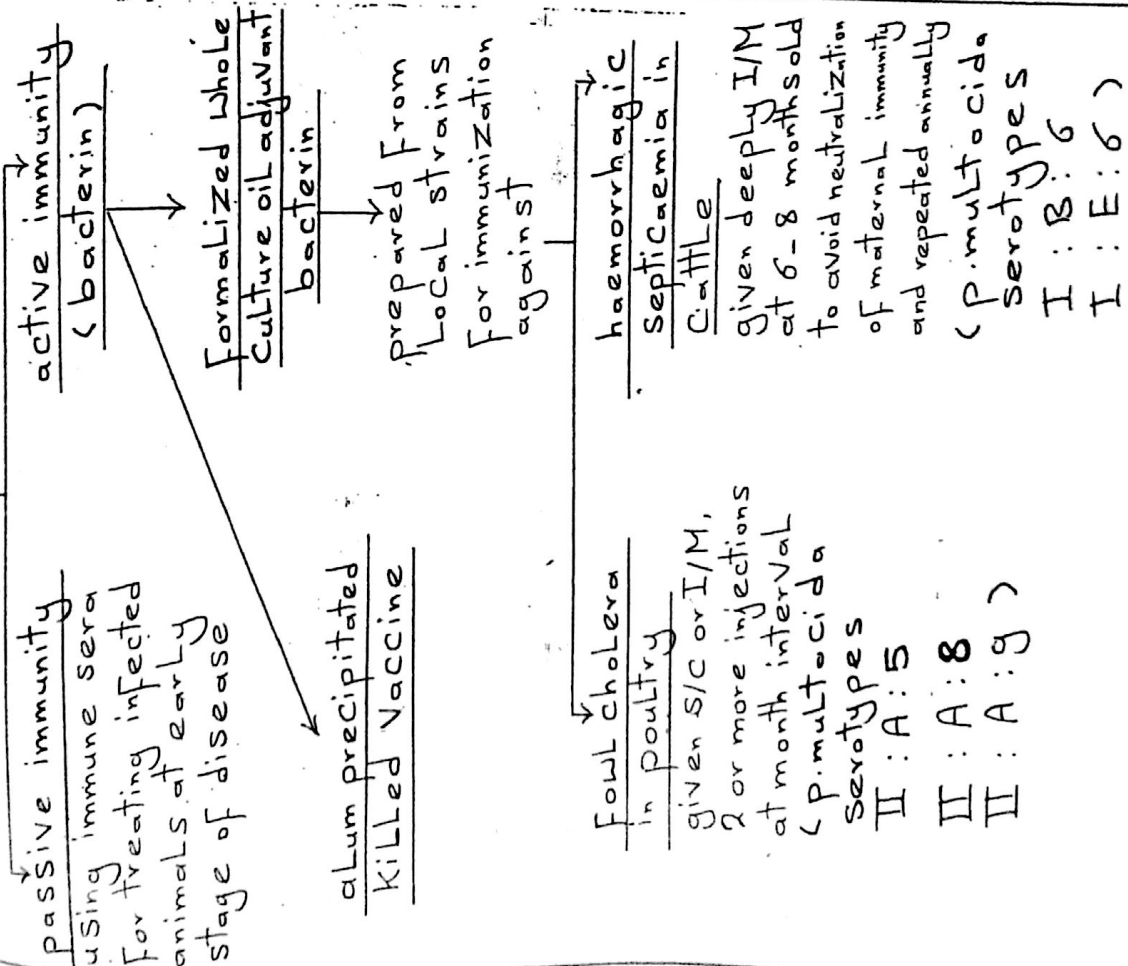
It is a live bovine strain that is attenuated by growth in potato-glycerin bile salts medium through several hundred subcultures

→ Massive immunization in animals is not recommended to avoid interference with Tuberculin test results

In man:

positive skin reactivity is developed in all BCG-vaccinated patients → thus skin testing can't be used to detect previous exposure to H. tuberculosis

Vaccination against Pasteurella multocida infection in buffaloes.



3) What are the virulence factors of ?

- 1- Streptococci.
- 2- Members of F Enterobacteriaceae.

4) What are the differences between..... ?

1- Motility of Listeriae and motility of Campylobacter species.

Motility of Listeriae	Motility of Campylobacter
characteristic tumbling motility (end-over-end) tumbling of individual cells with periods of quiescence) → when a 2-4 hrs broth culture incubated at 25°C and examined by the hanging drop technique.	characteristic darting movement (so, it is named artist's flying seagull organism) → seen by phase contrast or dark ground illumination microscope)
Listeriae are motile by a few (1-5) peritrichous flagella.	Campylobacter is motile by mono- and amphitrichous flagella

2- Fir tree growth appearance and invented one.

- Fir tree growth appearance: crail
all pathogenic clostridia except C. Colinum are positive with gelatin Liquefaction test (done by stabbing in gelatin shake agar) → show typical fir-tree growth in the bottom of the tube and no growth on the surface (as they are anaerobic)

Inverted Fir tree growth

B. anthracis in gelatin stab → show slow gelatin liquefaction producing an inverted Fir tree appearance (the line of stabbing is largest at the upper part of Culture)

3- Alpha and beta haemolysins of *S. aureus*. 2013 انظر

5) Write on :

1- Anton's test in rabbit.

Instillation of Few drops from the Culture of *L. monocytogenes* into the Conjunctival sac of rabbit or G. pig → Purulent Kerato-conjunctivitis appear within 24-36 hrs post-instillation.

2- Ascoli test. 2013 انظر

3- Antimicrobial resistance of *P. aeruginosa*.

P. aeruginosa is highly resistant to many antibiotics due to :

- ① Plasmid mediated resistance → involving
• modifying enzymes
• or change in outer membrane proteins (mutation of porin proteins in cell wall)

- ② aggregation of bacteria in vivo as microColonies inside a gelled ionized matrix → due to the production of exo-polysaccharide (extra-cellular Slime)

4- Cell wall composition of staphylococci.

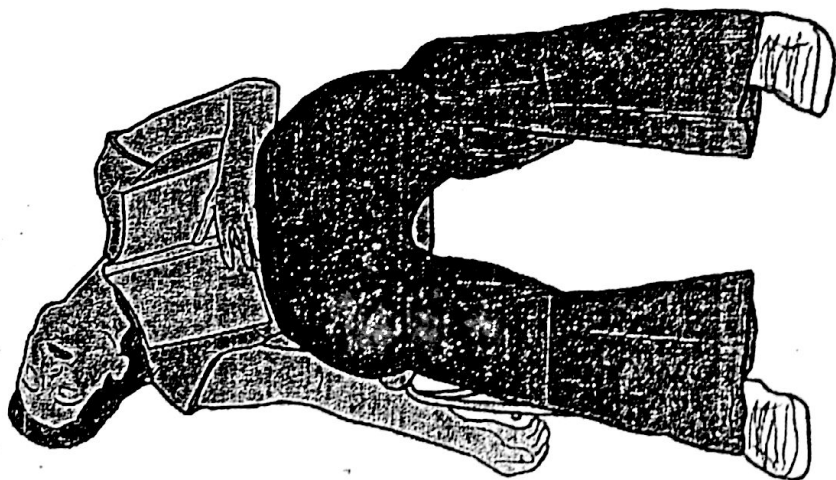
5- Types of pathogenic *E. coli*.

انظر
2013

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Systematic Bacteriology (Exams Guide)

امتحان ٢٠١٠



Bacteriology 2010

1) Some of pathogenic bacteria play a significant role as food poisoning organisms to human through consumption of meat and milk byproducts. Among them, some bacteria can live intracellular and resist phagocytic killing.

- 1-comment on this sentence demonstrating your answer with examples.
- 2-write a laboratory diagnosis of one of them transmitted by rodents. How can it resist intracellular killing?
- 3-which one lyses phagosome? Illustrate the mechanism by which lyses phagosome and attacks other host cells and discuss its general characters.

①

Food poisoning

Food infection
by bacteria itself

Food intoxication
by bacterial toxin

- e.g.
- Salmonella
 - E. coli
 - C. jejuni
 - Listeria
 - (L. monocytogenes and L. ivanovi)

- S. aureus
- B. cereus
- C. botulinum
- C. perfringens type A

②

Food Poisoning organism transmitted by rodents

Salmonella

Laboratory diagnosis of salmonella

1. Isolation:

1. Cultivation on selective enrichment broth:

- Such as
 - Selenite F broth
 - tetrathionate broth
 - Rappaport Vassiliadis broth

→ This step is done to enhance the growth of Salmonella and inhibit the growth of other enteric bacteria.

→ The inoculated tubes are incubated aerobically at 43°C or 37°C .

→ With addition of Na Sulphathiazole at 1.25 mg/L

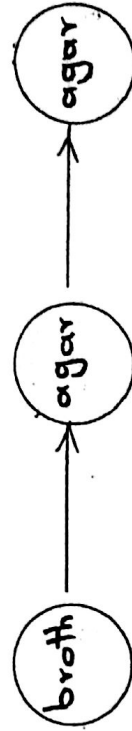
→ The incubation time → Not more than 18-20 hrs to suppress the growth of proteus spp.

2. Plating on selective differential solid media:

- Such as
 - MacConkey's agar
 - Salmonella-shigella agar (S-S agar)
 - Desoxycholate citrate agar (DCA)
 - brilliant green agar
 - xylose lysine desoxycholate agar (XLD)

- The inoculated plates are incubated aerobically at 37°C for 1-3 days.

- Suspected colonies of Salmonella are picked up and subcultured onto the surface of selective differential solid media to get a pure culture.



Q: How can you obtain a pure culture of an organism suspected to be salmonellae from intestinal contents (2006)

2. Identification:

a. Culture characters:

- on nutrient agar: Smooth colonies

- on MacConkey's agar:

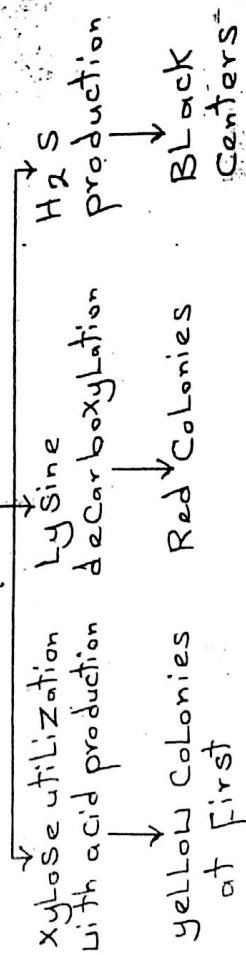
Salmonellae produce colorless colonies because they are non-lactose fermenter except S. arizonae (late lactose fermenter, after 3 days)

• on S-S agar: as MacConkey's agar

• on DCA: as MacConkey's agar

• on brilliant green agar:
pink colonies

• on XLD:



→ Indicator → phenol red indicator

b- Morphology:

- stain used → Gram's stain
- staining reaction → Gram -ve
- shape → rods
- size → small to medium
- spore → Non-Sporulated
- capsule → may be Capsulated
- motility → motile with peritrichous
Flagella except *S. pullorum* and *S. gallinarum* (Non-motile)

C-biochemical Identification:

H₂S urease I M V i C
+ - - + - +

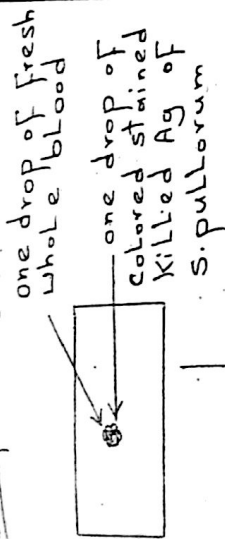
- gelatin Liquefaction test → -ve
- Reduce Nitrate to Nitrite
- do not ferment Sucrose, salicin and Lactose except *S. arizonae* which is Late Lactose fermenter.
- deCarboxylase LySine, arginine and ornithine.

d-Serological identification:

1-agglutination test:

Slide (rapid) agglutination test	Tube (slow) agglutination test
→ made by using polyvalent and monovalent O and H Salmonella antisera to determine the serovar of isolated strain.	It is a Confirmatory test for the slide test and used for determining the titre of infection.
→ must be confirmed by tube agglutination test.	

In poultry:



+ve result

visible clumping of colored antigen

→ this test is called

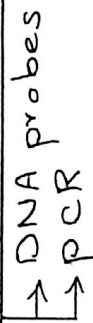
whole blood plate test

→ used in field.

2. other serological tests



e-Molecular techniques:



F-phage typing:

It is very important in epidemiology and based on the sensitivity of a particular isolate to a series of bacteriophages at appropriate dilutions.

• How Can Salmonella resist intracellular Killing?

1- Salmonellae are protected from acidic pH of phagosome by acid tolerance response (ATR) gene.

2- Catalase and superoxide dismutase → protect Salmonella from intracellular killing by phagocytes through their inhibition of oxidative killing inside phagocytic cells.

③ which one lyses phagosome? Illustrate the mechanism by which lyses phagosome and attacks other host cells and discuss its general characters.

Listeria

Mechanism:

L. monocytogenes induce phagocytosis through a membrane associated protein called internalin →

once ingested, the bacterium produce Listeriolysin (LLO) to

escape from the phagosome → then multiply rapidly in the

cytoplasm → move through the cytoplasm to invade adjacent

cells by polymerizing actin to form long tails.

• general characters of *Listeria*:

a - Morphology:

- Gram staining reaction → Gram +ve
- acid Fastness → Non-acid fast
- shape → Cocci bacilli
- size → small to medium
- arrangement → uncharacteristic
- Motility → motile by a few (1-5) peritrichous flagella (tumbling motility)
- Capsule → Non
- spore → Non

b - growth requirement:

- O₂ req. → Facultative anaerobes but growth is enhanced in presence of 10% CO₂ (Capnophilic)
- opt. temp. → 3 - 45°C (opt. 37°C)
(Cold enrichment)
- opt. pH → 5.6 - 9.6
- grow on nutrient agar but not on MacConkey's agar
- Can tolerate 10% NaCl (Halophilic).

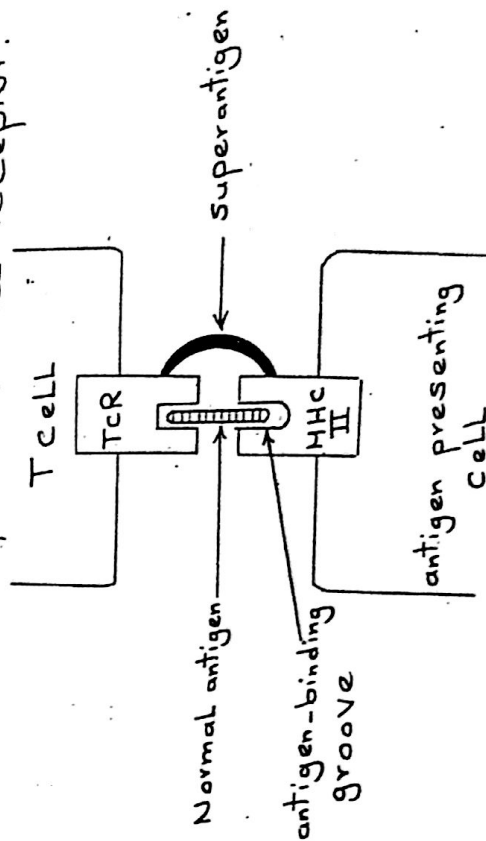
c - biochemically

- Catalase +ve
 - oxidase -ve
 - hydrolyze aesculin
- d - asymptomatic faecal carrier occur in man and many animals

2) Why?

1- Staphylococcal enterotoxins act as superantigens.

- Enterotoxins and TSST-1 are superantigens that may cause toxic shock syndrome.
- Superantigens stimulate T-cells non-specifically then cytokines are released in large amounts causing symptoms of toxic shock syndrome.
- Superantigen bind directly to MHC II on APC outside the conventional antigen binding groove. This complex recognizes only the Vβ element of the T-cell receptor.



Pathogenicity in laboratory animals must be done to P. multocida isolates.

because it is a commensal potential pathogen → present in upper resp. tract, when the resistance of A is lowered, it becomes active and produces a potent disease.

3- M-protein is important for pathogenicity of S. pyogenes.

- antiphagocytic (S. pyogenes and S. pyogenes)
- adhesin (S. equi subsp. equi)
- Cytotoxic to neutrophils.
- bind Factor H in Serum (regulatory protein for the alternative complement pathway) → thus, C3b that binds to the cell surface in the region of M-protein is degraded by Factor H.
- part of it is found in fimbriae → binding Fibrinogen from Serum → blocking the binding of complement to the underlying peptidoglycan → inhibit phagocytosis.
- Surface M-proteins contain antigenic epitopes → attach to heart muscle → leading to autoimmune rheumatic carditis (Rheumatic Fever).

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4- Cholesterol is required for isolation of mycoplasmas.

For formation of cell membrane which is a triple-layered and contains sterol (unit-membrane)

5- Killed brucella vaccines are mostly non-protective against infection.

because it produces humoral immunity but not cell-mediated immunity (which is protective)

3) Choose incorrect answer:

1- (E. coli, Brucella abortus, S. pyogenes, Pasteurella multocida)

..... is an obligate pathogen.

2- C. novyi differs from C. tetani in (motility, capsule, shape and position of spore, cell wall).

3- Lecithinase test is a diagnostic one for (S. aureus, S. agalactiae, Mycobacterium avium, toxicogenic strains of C. perfringens)

4- Bipolarity is a common character of (S. Typhimurium,

K. Pneumoniae, P. multocida, N. asteroides) Under microscope.

5- Lipoid waxy material in cell wall is a character of (micrococci, staphylococci, streptococci, mycobacteria, members of family Enterobacteriaceae)

6- Most strains of S. aureus are (sensitive, resistant) to (tetracyclines, gentamicin, penicillins, cephalosporins) due to the production of (staphylokinase, penicillinase, hyaluronidase, lecithinase) enzyme.

7-.....(peptidoglycan, M-protein, SPA) Is the cause of (pseudotuberculosis, immune, non-immune) reaction through its binding with (fab portion, FC portion, flagella, capsule, fimbriae) of immunoglobulins specially IgG one.

8-Glanders in equines is caused by (p. fluorescens, S. equi, B. mallei, R. equi) meanwhile anestrus in cows is caused by (brucellae, C. fetus subsp. venerealis, M. mycoides subsp. mycoides).

9-..... (ETEC, EIEC, EPEC, EHEC) attacks the intestinal epithelial cells causing (watery, bloody) diarrhea in contrast (ETEC, EIEC, EPEC) secretes (enterotoxins, cytotoxins, fibrinolysin) causing (food poisoning, haemorrhagic colitis) in human.

10-..... (haemolysins, endotoxins, siderophores, fimbriae, capsule) aid the bacteria in the competition with host for (folic acid, iron, sugar, protein). At the same time, bacteria produce (phosphatase, DNase, IROMPs, protease) responsible for the internalization of these compounds after binding with the required agent into bacterial cells.

4) How can you differentiate?

- 1- M. tuberculosis from M. bovis.
- 2- P. aeruginosa from P. fluorescens.
- 3- mycoplasma from ureaplasma and acheloplasma.

Differences between the 3 main types of Mycobacteria			
	M. tuberculosis	M. bovis	M. avium
Criteria	Human type	bovine type	avian type
1-Natural host	mainly human and primates	Cattle, human and others	Mainly birds but also pigs
2-opt. temperature	37°C	37°C	41-44°C
3-Incubation time	2-3 weeks	3-8 weeks	2-3 weeks
4-growth on nutrient agar	-	-	(good growth)
5-growth in glycerol broth	well growth with surface pellicle	poor growth	well growth with no pellicle
6-Colonies on dorset's egg medium	Eugenic (Luxuriant) Rough Dry	dysgenic (non-luxuriant) Smooth White	Eugenic (Luxuriant) Smooth moist and grayish
7-Morphology	Long, thin, curved and beaded	short and straight	pleomorphic
8-Biochemical test:			
• Niacin formation	+	-	-
• Nitrate reduction	+	-	-
• Tellurite reduction	-	-	+
9-pathogenicity to Lab. animals:			
• G. pig	strong	strong	-
• Rabbits	Mild	strong	Moderate
• Fowl	-	-	strong

Differences among pseudomonads and Burkholderiales

points of differences	B. mallei	B. pseudomallei	P. aeruginosa	P. fluorescens
① Motility	Non-motile	Motile by mono- or lophotrichous flagella	Motile by monotrichous flagella	Motile by lophotrichous flagella
② gelatin Liquefaction	-	+	+	+
③ growth at: 5°C (low temp) 42°C (high temp)	- -	- +	- +	+ -
④ pigment	brown	brown		
⑤ growth on MacConkey's agar	-	+	+	+
⑥ oxidation of Lactose	-	+	-	-
⑦ pathogenicity	glanders (equines)	Melioidosis (sheep)	variety of pathological conditions	pseudomonas disease in fish
⑧ diagnosis in living animal	Mallein Test	Melioidin Test	-	-
⑨ arginine dihydrolase Test	+	+	+	+
⑩ pH 16	+	+	-	-

3 - Mycoplasma from ureaplasma and acholeplasma

by **digitonin test** (sensitivity to digitonin)

Sensitive
(Inhibition of growth by digitonin)

Modified urease test

Resistant
(growth)
Acholeplasma
(Colonies upto 3 mm in diameter)

urease -ve

Mycoplasma
(Colonies 0.1 - 0.6 mm in diameter)

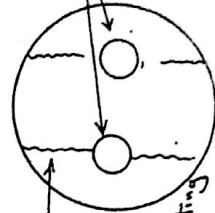
urease +ve

ureaplasma
(Colonies 0.01 - 0.05 mm in diameter "Tiny or T-mycoplasmas")

digitonin Sensitivity test:

→ used to differentiate between **Mycoplasma** and **acholeplasma** (non-pathogenic)

Lines of cultivation



Filter paper discs immersed in digitonin overnight in refrigerator (put on lines)

Incubate then examine under dissecting microscope.

IF no growth → Mycoplasma (sensitive to digitonin)

Modified urease test:

aim:
biochemical test used for differentiation between Mycoplasma and ureaplasma.

Technique:

Mixture of equal parts of 10% urea soln. + 0.8% manganese chloride → apply directly onto 40 hrs old microcolonies



+ve result

Immediate colour reaction goes from light to dark brown and finally to black (due to deposition of manganese on the surface of colonies).

+ve result → ureaplasma
-ve result → Mycoplasma

5-Discuss briefly:

- 1- immunization against B. anthracis.
- 2- Dot . ELISA test for diagnosis of B. mallei.

ذکر کنید

6) True or false and explain

periplasmic flagella

لا

1- spirochaetes are motile organisms with polar flagella. X

2- spores of C. ovis are found subterminal. X

as C. ovis are non-spore forming

3- septic shock syndrome is caused by S. aureus when it reaches the blood stream. ✓

due to massive complement activation induced by SPA-bound immunoglobulin and exposed peptidoglycan.

4- pathogenicity of C. botulinum depends on its invasion to host cells. X

as C. botulinum is non-invasive Mo that have no power to invade living tissues and their pathogenicity depends on production of highly powerful neurotoxin in contaminated canned or salted fish.

5- Most of staphylococci are sensitive to bacteriophages. ✓

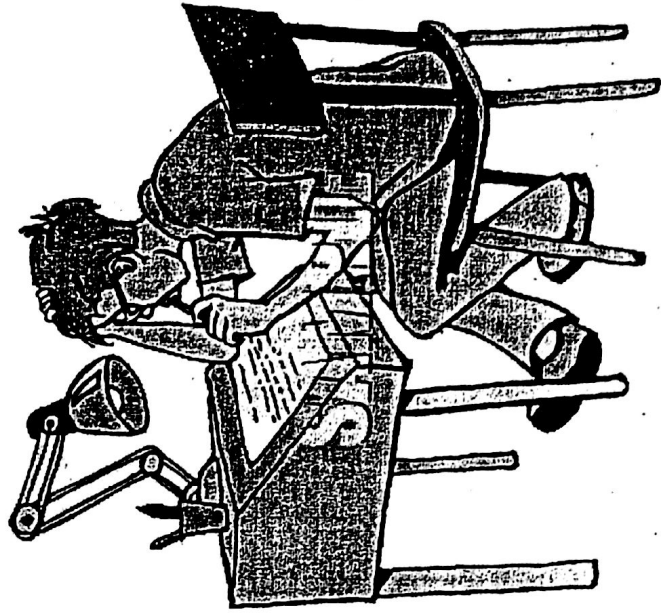
due to presence of teichoic acid in their cell wall which is the site of bacteriophage adhesion.

6- CAMP test is a character of N. asteroides.

Systematic Bacteriology

(Exams Guide)

امتحان ٢٠٠٩



C. jejuni is the cause of contagious bovine pleuropneumonia. X

Mycoplasma mycoides subsp. mycoides

8-Lancefield classification of streptococci depends on haemolysis on blood agar. X

C-antigen in their cell wall

9-S. aureus has the ability to clots oxalated or citrated plasma. ✓

due to production of coagulase enzyme

10-Ascoli test is used for detection of Brucella antibodies in sera of infected animals. X

Bacillus anthracis in dead animals

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Bacteriology 2009

1- some of pathogenic bacteria to farm animals play a

significant role as occupational diseases in human:

a- comment on this sentence demonstrating your answer with examples

b- write a laboratory diagnosis of one of them causing infertility in male animals

c- discuss vaccination of one of them causing sudden death in sheep

d- illustrate the allergic test of one of them depending on delayed type of hypersensitivity and can be applied on both human and animals.

a - bacteria causing occupational diseases in human

- B. anthracis
- B. abortus
- Mycobacterium tuberculosis

b - Lab. diagnosis of brucella

c - Vaccination against B. anthracis

d - Tuberculin test

2- Why?

a- P. Pyocyanus is immunosuppressive organism

① produce proteases → 2 types
 • Elastase → cleave IgG, IgA and Complement.
 • Elastase and alkaline protease together → inactivate IFN γ and TNF → suppress phagocytosis.

② produce Cytotoxin → cytotoxic for most eukaryotic cells including neutrophils.

③ produce exotoxins → 2 types

• Exotoxin-A → toxic for monocytes and bone marrow

• Exotoxin-S → interfere with phagocytic killing.
 ④ capsule → antiphagocytic

b- salmonellae resist intracellular killing inside phagocytes

ذكر قبل

c- ONPG test is used for diagnosis of S. arizonae

because it is late lactose fermenter

d- M. haemolytic needs blood for their growth

because it is fastidious H.O

e- L. monocytogenes forms long tail inside phagocytes

ذكر مرفق

3) Choose the correct answer and treatment

1- (Salmonella Enteritidis , Brucella abortus , Mycobacterium tuberculosis , Pasteurella Multocida) Is a commensal potential pathogen.

2- ETEC differs from EIEC in (Motility, Capsule, enterotoxins production , microscopic appearance)

3- CAMP Test is a diagnostic one for (S. aureus, S. agalactiae, Mycobacterium avium, Brucella melitensis)

4- Pitting of loffler's serum slope is character of (A. bovis, A. Pyogenes, C. ovis) depending on (Lipolytic Enzymes, saccharolytic enzymes, Proteolytic enzymes).

5- Teichoic acid in cell wall is a character of (micrococci, staphylococci, streptococci, mycobacteria, members of family Enterobacteriaceae).

4. Tabulate:

1. biochemical differences among E. coli, Salmonellae and Klebsiellae

Test	E. coli	Klebsiella	Salmonella
H ₂ S	-	-	+
urease	-	slow	-
IMViC	++--	--++	+-+-

2. Differences between P. multocida and M. haemolytica.

3. Differences between C. ovis and R. equi.

ذكره قبل

5. Discuss:

1- antigenic structure of pasteurellae and immunization against them.

2- Recent taxonomic classification of mycoplasmas.

under phylum (division) Firmicutes due to low G-C content (18-40 mol%) small genome

ذكره قبل

5. Put the mark (✓) or (X) in front of each of the following sentences and explain your answer (please, no marks without explain):

1- There is a strong relationship between motility of C. perfringens and its pathogenicity. X as it is non-motile

2- Spores of S. aureus are found subterminal. X as it is non sporulated

3- tetanospasmin is produced only by all toxigenic strains of C. tetani. X as it is produced by all toxigenic and non-toxigenic strains

4- It is so simple and easy to prepare neutralizing antitoxins against C. botulinum toxins. X because they are 8 antigenically distinct exotoxins

5- Symptomatic anthrax is a disease infecting sheep and caused by C. perfringens type A through ingestion of its spores. X caused by C. chauvoei through wound infection by its spores.

6- Bacillary haemoglobinuria in cattle is a disease caused by Borrelia anserina. X clonov type D

7- Lecithinase test is one of the tests which must be done on blood agar X egg yolk

8- Rabbits and pigeons are susceptible to infection by C. tetani. X resistant

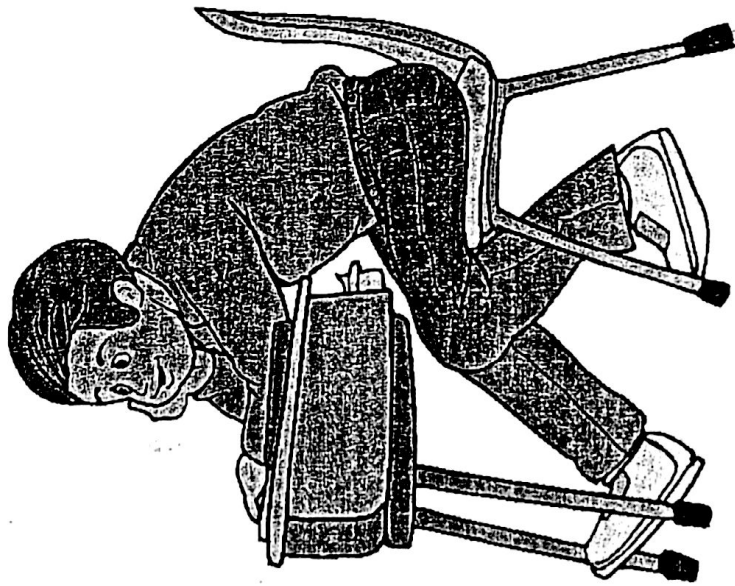
9- Pathogenicity of clostridia species are mostly correlated with gelatin hydrolysis. X toxin production

10- Both acid and formalin stability are two important characters of C. tetani. X

Tetanospasmin of C. tetani is both acid and formalin labile.

Systematic Bacteriology (Exams Guide)

امتحان ٢٠٠٨



Bacteriology 2008

1- Pleomorphism is one of important characters of pathogenic microorganism. some of them are originally pleomorphic, others are pleomorphic after prolonged incubation:

a- define pleomorphism demonstrating your answer with examples and explain the causes of pleomorphism in each case.

pleomorphism:

change bacterial morphology under microscope examples:

↓
originally pleomorphic organism

Mycoplasma
(due to Lack of cell wall)

↓
organisms pleomorphic after prolonged incubation

- P. multocida
- brucella
- M. avium
- Corynebacterium
- actinomyces
- NoCardia
- Rhodococcus

b-write on:

- a- General characters of an originally pleomorphic one. → Mycoplasma
- b- Laboratory diagnosis of one becomes pleomorphic after prolonged incubation. → Lab. diagnosis of P. multocida
- c- Vaccination against any one of them. → Vaccination of brucella

Which type of microorganism produces each of the following products:

- 1-LLO. → *L. monocytogenes*
 2-Coagulase. → *S. aureus*
 3-pyocyanin. → *P. aeruginosa*
 4-Tetanus toxin. → *C. tetani*
 5-Lecithinase → *Clostridium C. perfringens* type A)

III-where each of the following components is found, which type or microorganism(s) contain it and, what is its significance?

- 1-Mycolic acid
 2-C-Antigen.
 3-SPA.
 4-Polypeptide capsule.
 5-Metachromatic granules.

Component	M.O	Significance
1-Mycolic acid	<i>Mycobacteria</i> (cell wall)	• resp. for acid fastness. • virulence • prevent attack of mycobacteria in the phagocytic granule • protect ext. mycobacteria from complement deposition
2-C-antigen	<i>Streptococci</i> (cell wall)	Lancefield classification
3-SPA	<i>S. aureus</i> (cell wall)	• pseudoimmune reaction. • septic shock syndrome
4-polypeptide capsule	<i>B. anthracis</i>	Virulence
5-Metachromatic granules	1- <i>C. diphtheriae</i> 2- <i>B. mallei</i> 3- <i>P. multocida</i>	diagnostic feature → Bipolarity

4) Choose the correct complementary answer from group B to the sentence in group A ?

(5 marks)

Group A	Group B
1. <i>P. aeruginosa</i> is highly <u>⑤</u> of Runyon's classification.	1. proteolytic and saccharolytic organism.
2. mycobacteria depends on <u>⑥</u> .	2. two chromosomes.
3. Pitting of Löffler's serum slope occurs <u>⑪</u> .	3. <i>S. agalactiae</i> .
4. <i>Corynebacteria</i> are included recently in <u>⑫</u> .	4. resistant to the effect of heat.
5. <i>Brucella</i> species contain <u>②</u> .	5. resistant to a wide range of chemotherapeutic agents.
6. CAMP test is positive with <u>③</u> .	6. rate of growth and pigment production.
7. Presence of oxalates in culture media <u>⑩</u> .	7. fab portion of antibody causing pseudo-immune reaction.
8. <i>L. monocytogenes</i> is regarded as <u>⑭</u> .	8. with <i>A. bovis</i> .
9. Colonies of bovine type of mycobacteria are <u>⑬</u> .	9. enhances the production of <i>B. anthracis</i> spores.
10. <i>C. perfringens</i> is a <u>⑬</u> .	10. the division Actinobacteria.
	11. stabbing in soft agar.
	12. dysgonic in their growth.
	13. produced in a large amount and less potent in their action.
	14. one of halophilic organisms.
	15. the division Gramicutes.
	16. motile with axial filament.
	17. diagnostic for <i>Brucella</i> infection.

GOOD LUCK

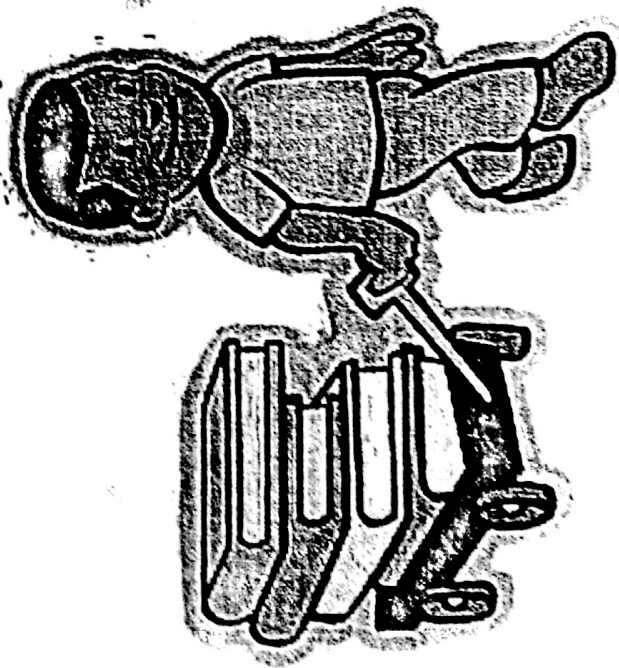
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Systematic Bacteriology

COLLECTIONS



pleomorphism:

def. Change morphology of M.O under microscope

→ organism originally pleomorphic in its nature

→ Mycoplasma (Lack cell wall) the smallest one causing pneumonia in Cattle.

→ organism pleomorphic after repeated subculture or prolonged incubation

- 1- pasteurella
- 2- brucella
- 3- Mycobacterium avium
- 4- Corynebacterium
- 5- actinomyces
- 6- Nocardia
- 7- Rhodococcus

Facultative intracellular bacteria:-

- 1- Mycoplasma
- 2- Salmonella
- 3- brucella
- 4- Mycobacterium (T.B)
- 5- Corynebacterium ovise
- 6- Listeria
- 7- Nocardia

- Its control is difficult
- produce asymptomatic carrier

Why their control is difficult?

Because they are facultative intracellular bacteria (resist intracellular killing by phagocytes). So, infected cases can be converted into carrier state even after treatment because antibiotics affect only extracellular bacteria not intracellular one.

Bacteria Cause obligatory infection (obligate pathogen):

its presence in the host's body is accompanied by disease production such as →

- 1- Mycobacterium → affect lung and its associated Ln
- 2- Brucella → Cause storm of abortion in cattle
- 3- Salmonella → Cause food poisoning (infection) in man
- 4- B. anthracis → Cause sudden death in sheep
- 5- B. mallei → Cause glanders in equine

(2)

Food poisoning bacteria:

Food intoxication
Caused by bacterial toxin

- | | |
|---------------------------|-------------------------|
| 1- S. aureus | Food infection |
| 2- Cl. botulinum | Caused by bacteria |
| 3- Cl. perfringens Type A | 1- Salmonella |
| 4- Bacillus cereus | 2- Campylobacter jejuni |
| | 3- E. coli |
| | 4- Listeria |

N.B:

- Cl. botulinum → release neurotoxins in contaminated salted fish causing botulism (food intoxication).
- Salmonella → Facultative intracellular → transmitted by rodents
- Listeria → Facultative intracellular (lyse phagosome and attack other host cells)

Bacteria Causing occupational diseases in humans:

- 1- Brucella → Cause storm of abortion in cattle
- 2- B. anthracis → Cause infertility in animals
- 3- B. mallei → Cause sudden death in sheep
- Cause lymphangitis in horse
- diagnosed by Mallein test

(3)

4- Mycobacterium → diagnosed by tuberculin test (allergic test depend on delayed type of hypersensitivity and can be applied on both human and animals)

Bacteria have metachromatic granules:

- 1- Burkholderia mallei
- 2- Corynebacterium diphtheriae
- 3- Pasteurella multocida (bipolarity)

Bacteria causing strauss reaction:

- 1- Burkholderia mallei
- 2- Brucella abortus
- 3- Corynebacterium ovis

Pyogenic M.O:

- 1- Staphylococcus aureus
- 2- Some of Streptococcaceae (e.g. S. pyogenes)
- 3- Corynebacterium
- 4- Neisseria
- 5- Rhodococcus
- 6- Actinomyces pyogenes
- 7- Pseudomonas aeruginosa

(4)

Good Luck !

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